16jan03 15:54:30 User217743 Session D593.2
$0.00 0.072 DialUnits File410
$0.00 Estimated cost File410
$0.04 TELNET
$0.04 Estimated cost this search
$0.05 Estimated total session cost 0.244 DialUnits
File 155:MEDLINE(R) 1966-2002/Dec W5
*File 155: Updating of completed records has resumed.See Help News155. Alert feature enhanced with customized scheduling. See HELP ALERT.

Set Items Description
--- ----- -----------
? s gamma()globin and hemin
188915 GAMMA
9468 GLOBIN
873 GAMMA(W)GLOBIN
2903 HEMIN
S1 54 GAMMA()GLOBIN AND HEMIN
? s s1 and induction
54 S1
222499 INDUCTION
S2 21 S1 AND INDUCTION
? s s2 and assay
21 S2
306236 ASSAY
S3 2 S2 AND ASSAY
? t s3/3,ab/1,2

3/3,AB/1
DIALOG(R)File 155:MEDLINE(R)

10989439 20572538 PMID: 11121491
Quantitative analysis of globin gene *induction* in single human erythroleukemic cells.
Smith R D; Malley J D; Schechter A N
Laboratory of Chemical Biology, National Institute of Diabetes and Digestive and Kidney Diseases and Computational Bioscience and Engineering Laboratory, Center for Information Technology, National Institutes of Health, Bethesda, MD 20892, USA.
Nucleic acids research (ENGLAND) Dec 15 2000, 28 (24) p4998-5004, ISSN 1362-4962 Journal Code: 0411011
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
The mechanisms involved in the normal developmental regulation of globin gene expression, and the response to pharmacological agents that elevate fetal hemoglobin, may be expected to involve either changes in each cell or a selection process affecting subsets of differentiating erythroid cells. To study these mechanisms we have developed assays to measure mRNA levels in single erythroid cells. The *assay* involved the use of globin-specific probes, with no detectable cross-reactivity, in real-time, fluorescence-based quantitative PCR (Q-PCR). We had previously used this Q-PCR method to measure globin mRNA levels in cultures of primary erythroid cells demonstrating that drugs like hydroxyurea, 5-azacytidine and butyric acid each yielded increases in gamma/(gamma + ss) mRNA ratios, with differential effects on ss-globin levels. We have now extended this approach to measure globin mRNA levels in single K562 cells, a human erythroleukemic cell line, with and without 30 &mgr;M *hemin* treatment. *Hemin* exposure increases total hemoglobin levels by approximately 9-fold and total alpha-, varepsilon- and *gamma*- *globin* mRNA levels by 1.5-2.3-fold. Single cell analyses showed initial wide distributions of each of the three individual globin mRNA levels with most cells having detectable but very low levels of each globin transcript. *Hemin* *induction* shifted the distributions to higher levels, with a tendency to residual left skewing as some cells remained with very low expression levels despite the effect of *hemin* in increasing expression in most of these low expressing cells. Thus transcriptional heterogeneity remains a crucial variable, even in this extensively used model of human erythroid biology, and clearly influences strongly the response to inducing agents. These methods may enable us to define better possible molecular and/or cellular models of globin gene modulation.

3/3,AB/2
DIALOG(R)File 155:MEDLINE(R)

09958279 98403713 PMID: 9734644
Factor binding to the human *gamma*-*globin* gene distal CCAAT site: candidates for repression of the normal gene or activation of HPFH mutants.
Partington G A; Patient R K
Developmental Biology Research Centre, The Randall Institute, Division of Biomedical Sciences, King's College London.
British journal of haematology (ENGLAND) Sep 1998, 102 (4) p940-51, ISSN 0007-1048 Journal Code: 0372544
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
We have examined factor binding to the distal human *gamma*- *globin* CCAAT site and three naturally occurring hereditary persistence of fetal haemoglobin (HPFH) mutations of this site. Factor binding was examined using nuclear extracts from the erythroleukaemic cell lines K562 and MEL, and from A4 cells, a non-transformed mouse bone marrow stem cell line, using the electrophoretic mobility shift *assay*. Under standard binding conditions, in addition to the

previously reported binding by a CCAAT factor (CP1) and GATA-1, the wild-type (wt) sequence bound high mobility factors which appeared to be GATA-2 isoforms. However, when the non-specific competitor conditions were varied, the binding profile with K562, but not MEL nuclear extract, was substantially altered. CP1 and GATA-1 were absent, and two new factors were detected, one of which bound preferentially to the Greek and Japanese non-deletion HPFH mutants. However, binding by the GATA-2 isoforms to the wt sequence was maintained with both cell types, as it was using the A4 cell line. With modified binding conditions, in A4 cells the two non-deletion and the Black deletion HPFH mutants each had a different protein binding profile which was lost on erythroid *induction* of the cells. We discuss the possibility that the GATA-2 isoforms bound to the wt sequence may function to suppress wt gamma gene expression in the bone marrow. Additionally, those factors which bind preferentially either to the deletion or non-deletion HPFH mutants may play positive roles in establishing an active chromatin structure. ? *******ds
? ds

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Set    Items  Description
S1        54  GAMMA()GLOBIN AND HEMIN
S2        21  S1 AND INDUCTION
S3         2  S2 AND ASSAY
? s fetal()hemoglobin
      193735  FETAL
       51179  HEMOGLOBIN
    S4   3977  FETAL()HEMOGLOBIN
? s s4 and hemin
        3977  S4
        2903  HEMIN
    S5     41  S4 AND HEMIN
? s s5 and assay
          41  S5
      306236  ASSAY
    S6      4  S5 AND ASSAY
? s s6 not s3
           4  S6
           2  S3
    S7      2  S6 NOT S3
? t s7/3,ab/1,2

 7/3,AB/1
DIALOG(R)File 155:MEDLINE(R)

05828801  88250918  PMID: 3382158
 Quantitation of hemoglobin with the Vision Analyzer by use of the alkaline hematin reaction.
 Pesce M A; Giacomo D F
 Columbia Presbyterian Medical Center, Special Chemistry Laboratory, New York, NY 10032.
 Annals of clinical and laboratory science (UNITED STATES) Mar-Apr 1988, 18 (2) p168-73, ISSN 0091-7370 Journal Code: 0410247 Document type: Journal Article
 Languages: ENGLISH
 Main Citation Owner: NLM
 Record type: Completed
 Blood is drawn into capillary tubes containing saponin and the tubes placed into the reagent packs. Hemoglobin is denatured by mixing the hemosylate with a reagent containing lithium hydroxide and a non-ionic detergent. The absorbance is measured bichromatically at wavelengths of 577 and 633 nm. The calibration curve is stable and can be stored for at least 30 days. There are no interferences from *fetal* *hemoglobin*, glycosylated hemoglobin (20 percent), hemoglobin S, samples with hematocrits up to 0.55, paraproteins, and lipemia. Specimens with rouleau formation, nucleated and fragmented red blood cells, target cells, ovalocytes, teardrop cells, spherocytes, leukocyte counts of 29 X 10(9) per L and reticulocyte counts of 0.32; Howell-Jolly bodies did not interfere with the *assay*. The within run and between run precision gave average coefficient or variations of 2.3 and 1.9 percent, respectively. Comparison of the hemoglobin results obtained in 149 samples with the Vision (y) and Coulter Counter System (x) gave r = 0.987, Y = 1.01X - 1.89 g per L.

 7/3,AB/2
DIALOG(R)File 155:MEDLINE(R)

03882243  82157192  PMID: 6175208
  Identification and quantitation of embryonic and three types of *fetal* *hemoglobin* produced on induction of the human pluripotent leukemia cell line K-562 with *hemin*.
 Fuhr J E; Bamberger E; Lozzio C B; Lozzio B B; Felice A E; Altay G; Webber B B; Reese A L; Mayson S M; Huisman T H
 American journal of hematology (UNITED STATES) Feb 1982, 12 (1) p1-12, ISSN 0361-8609 Journal Code: 7610369
 Contract/Grant No.: CA 18185-06; CA; NCI; HLB 23736; HL; NHLBI; HLB-05168 ; HL; NHLBI; +
 Document type: Journal Article
 Languages: ENGLISH
 Main Citation Owner: NLM
 Record type: Completed
 The hemoglobins synthesized by the pluripotent K-562 leukemia cell line of human origin after induction with *hemin* have been isolated by DEAE-cellulose chromatography and characterized by electrophoresis, high pressure liquid chromatography, and a radioimmunological *assay*. Six hemoglobin zones have been observed with the following likely compositions. Zone 1: alpha 2 epsilon 2, or HB Gower-2; zone 2: zeta 2
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epsilon 2, or HB Gower-1; zone 3: zeta 2 gamma 2, or HB Portland-I; zone 4: Hb F, or alpha 2 gamma 2; zone 5: a mixture of acetylated HB Portland-I and Hb F; zone 6: Hb Bart's, or gamma 4. The embryonic Hbs (zones 1, 2, and 3) constituted 50%-75% of the total Hb present; the quantities varied from one experiment to the other. Both Hb Gower-1 and Hb Gower-2 were present. The gamma chain was heterogeneous and contained the G gamma, A gamma I, and A gamma T types in a ratio of about 4:2:1, indicating a heterozygosity for the Ile leads to Thr substitution at position gamma 75. The methodology used can be applied for additional studies evaluating quantitative changes in Hb types due to in vitro manipulations.

? ds

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Set    Items  Description
S1        54  GAMMA()GLOBIN AND HEMIN
S2        21  S1 AND INDUCTION
S3         2  S2 AND ASSAY
S4      3977  FETAL()HEMOGLOBIN
S5        41  S4 AND HEMIN
S6         4  S5 AND ASSAY
S7         2  S6 NOT S3
? s s1 and s4
         54 S1
       3977 S4
    S8    15 S1 AND S4
? s s8 not s6
         15 S8
          4 S6
    S9    13 S8 NOT S6
? t s9/3,ab/all
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9/3,AB/1
DIALOG(R)File 155:MEDLINE(R)

14037796 22320940 PMID: 12393613

Development of sensitive fluorescent assays for embryonic and *fetal* *hemoglobin* inducers using the human beta -globin locus in erythropoietic cells.

Vadolas Jim; Wardan Hady; Orford Michael; Voullaire Lucille; Zaibak Faten ; Williamson Robert; Ioannou Panayiotis A; et al

Cell and Gene Therapy Research Group, The Murdoch Children's Research Institute, Royal Children's Hospital, Melbourne, Australia; and the Cyprus Institute of Neurology and Genetics, Nicosia, Cyprus.

Blood (United States) Dec 1 2002, 100 (12) p4209-16, ISSN 0006-4971 Journal Code: 7603509

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: In Process

Reactivation of *fetal* *hemoglobin* genes has been proposed as a potential therapeutic procedure in patients with beta-thalassemia, sickle cell disease, or other beta-hemoglobinopathies. In vitro model systems based on small plasmid globin gene constructs have previously been used in human and mouse erythroleukemic cell lines to study the molecular mechanisms regulating the expression of the fetal human globin genes and their reactivation by a variety of pharmacologic agents. These studies have led to great insights in globin gene regulation and the identification of a number of potential inducers of *fetal* *hemoglobin*. In this study we describe the development of enhanced green fluorescence protein (EGFP) reporter systems based on bacterial artificial chromosomes (EBACs) to monitor the activity of the epsilon-, (G)gamma-, (A)gamma-, delta-, and beta-globin genes in the beta-globin locus. Additionally, we demonstrate that transfection of erythroleukemia cells with our EBACs is greatly enhanced by expression of EBNA1, which also facilitates episomal maintenance of our constructs in human cells. Our studies in human cells have shown physiologically relevant differences in the expression of each of the globin genes and also demonstrate that *hemin* is a potent inducer of EGFP expression from EGFP-modified epsilon-, (G)gamma-, and (A) *gamma*-*globin* constructs. In contrast, the EGFP-modified delta- and beta-globin constructs consistently produced much lower levels of EGFP expression on *hemin* induction, mirroring the in vivo ontogeny. The EGFP-modified beta-globin eukaryotic BAC (EBAC) vector system can thus be used in erythroleukemia cells to evaluate induction of the epsilon- and *gamma*-*globin* genes from the intact human beta-globin locus.

9/3,AB/2
DIALOG(R)File 155:MEDLINE(R)

11110969 21117126 PMID: 11172039

Mechanism for fetal globin gene expression: role of the soluble guanylate cyclase-cGMP-dependent protein kinase pathway.

Ikuta T; Ausenda S; Cappellini M D

Center for Human Genetics, Boston University School of Medicine, Boston, MA 02118, USA. tikuta@bu.edu

Proceedings of the National Academy of Sciences of the United States of America (United States) Feb 13 2001, 98 (4) p1847-52, ISSN 0027-8424 Journal Code: 7505876

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Despite considerable concerns with pharmacological stimulation of *fetal* *hemoglobin* (Hb F) as a therapeutic option for the beta-globin disorders, the molecular basis of action of Hb F-inducing agents

remains unclear. Here we show that an intracellular pathway including soluble guanylate cyclase (sGC) and cGMP-dependent protein kinase (PKG) plays a role in induced expression of the *gamma*-*globin* gene. sGC, an obligate heterodimer of alpha- and beta-subunits, participates in a variety of physiological processes by converting GTP to cGMP. Northern blot analyses with erythroid cell lines expressing different beta-like globin genes showed that, whereas the beta-subunit is expressed at similar levels, high-level expression of the alpha-subunit is preferentially observed in erythroid cells expressing *gamma*- *globin* but not those expressing beta-globin. Also, the levels of expression of the *gamma*-*globin* gene correlate to those of the alpha-subunit. sGC activators or cGMP analogs increased expression of the *gamma*-*globin* gene in erythroleukemic cells as well as in primary erythroblasts from normal subjects and patients with beta-thalassemia. Nuclear run-off assays showed that the sGC activator protoporphyrin IX stimulates transcription of the *gamma*-*globin* gene. Furthermore, increased expression of the *gamma*-*globin* gene by well known Hb F-inducers such as *hemin* and butyrate was abolished by inhibiting sGC or PKG activity. Taken together, these results strongly suggest that the sGC-PKG pathway constitutes a mechanism that regulates expression of the *gamma*-*globin* gene. Further characterization of this pathway should permit us to develop new therapeutics for the beta-globin disorders.

9/3,AB/3
DIALOG(R)File 155:MEDLINE(R)

09636671 98062479 PMID: 9398338
In vivo footprinting using N-ethyl,N-nitrosourea: improved resolution of the DNA-protein interactions in the human *gamma*-*globin* gene promoter region.
Kim I H; Rodgers G P
Laboratory of Chemical Biology, National Institutes of Health, Bethesda, Maryland 20892, USA.
Analytical biochemistry (UNITED STATES) Dec 1 1997, 254 (1) p1-8, ISSN 0003-2697 Journal Code: 0370535
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
N-Ethyl, N-nitrosourea (ENU) was used as a probing agent in conjunction with a modified ligation-mediated polymerase chain reaction in a new in vivo footprinting procedure. In the present work, we examined the promoter region of the human *gamma*-*globin* gene under both uninduced and *hemin* -induced conditions in K562 cells. In the course of comparing this method with the standard dimethyl sulfate (DMS) in vivo method and previously reported results, we were able to verify our new method. However, discrepancies between these methods were observed at the stage selector element, -50 region, of *gamma*-*globin* promoter. Our in vivo footprinting result showed DNA-protein interaction at this region under the *hemin* -induced condition which was not revealed by the conventional DMS in vivo footprinting method. This approach, using ENU-modified in vivo footprinting, is now being applied to clarify the mechanism of cytotoxic drug-induced *fetal* *hemoglobin* augmentation. Copyright 1997 Academic Press.

9/3,AB/4
DIALOG(R)File 155:MEDLINE(R)

09482546 97390440 PMID: 9242673
Globin gene silencing in primary erythroid cultures. An inhibitory role for interleukin-6.
Ferry A E; Baliga S B; Monteiro C; Pace B S
Department of Structural and Cellular Biology, University of South Alabama, Mobile, Alabama 36688, USA.
Journal of biological chemistry (UNITED STATES) Aug 8 1997, 272 (32) p20030-7, ISSN 0021-9258 Journal Code: 2985121R
Contract/Grant No.: HL 38639-09; HL; NHLBI
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
There are numerous similarities between the erythroid and megakaryocytic lineages which suggest that commitment to either lineage occurs relatively late in hematopoiesis. Commitment toward megakaryocyte development requires obligatory silencing of erythroid-specific genes. Therefore, we investigated the effects of interleukin-6, a known inducer of thrombocyte production, on globin gene expression during erythroid differentiation. Studies in K562 cells demonstrated inhibition of *gamma* *globin* gene mRNA production and chain biosynthesis in the presence of exogenous interleukin-6 which was abrogated by anti-interleukin-6 monoclonal antibody. Similar studies in primary erythroid progenitors showed inhibition of burst-forming unit-erythroid colony formation when interleukin-6 was added late in cultures with decreased gamma and beta globin gene mRNA production. Protein binding studies demonstrated an increase in activator protein-1 binding to its consensus sequence by 24 h of interleukin-6 treatment. Inhibition of activator protein-1 gene activity had no effect on gamma gene silencing by interleukin-6. A potential interleukin-6 response element was identified in

the *gamma* *globin* gene. Interleukin-6 treatment led to a rapid increase in protein binding to the target DNA sequence. These results suggest that interleukin-6 may play an important role in globin gene silencing during megakaryocytic lineage commitment.

9/3,AB/5
DIALOG(R)File 155:MEDLINE(R)

09479054 97364261 PMID: 9220539
Modulation of globin gene expression in cultured erythroid precursors derived from normal individuals: transcriptional and posttranscriptional regulation by *hemin*.
Kollia P; Noguchi C T; Fibach E; Loukopoulos D; Schechter A N First Department of Medicine, University of Athens, Laikon General Hospital, Greece.
Proceedings of the Association of American Physicians (UNITED STATES) Jul 1997, 109 (4) p420-8, ISSN 1081-650X Journal Code: 9514310 Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
We are interested in the genetic mechanisms whereby several classes of drugs increase *fetal* *hemoglobin* (HbF) in patients with sickle-cell anemia or beta-thalassemia. Recently, we have shown (Kollia et al., Proc. Natl. Acad. Sci. U.S.A. 93: 5693, 1996) that cultured primary human adult erythroid cells (hAEC) offer a useful model for the study of transcriptional and posttranscriptional regulation of globin gene expression. We have found also that *hemin* markedly increases HbF levels in these cells. We report here the effect of *hemin* on globin gene transcription and RNA processing in hAEC. Quantitative reverse transcriptase-polymerase chain reaction analysis showed that the *gamma*-*globin* message levels in the cytoplasm and nucleus were increased two-fold by *hemin* . In the untreated cells, only spliced gamma-transcripts were detected in the cytoplasm, indicating that only completely processed gamma-RNA is transported to the cytoplasm, whereas approximately half of the nuclear *gamma*-*globin* RNA transcripts were unspliced. After treatment with *hemin* , correctly spliced gamma-transcripts increased in the cytoplasm and nucleus, while the unprocessed gamma-transcripts decreased in number in the nucleus. We also studied epsilon-globin RNA transcripts; in the cytoplasm of untreated cells, only correctly processed transcripts were present, whereas the nuclear epsilon-globin RNA transcripts were unspliced. In *hemin* -induced cells, unspliced nuclear epsilon-transcripts decreased in number. In contrast to the gamma- and epsilon-globin genes, the levels of full-length, correctly spliced beta-globin message are not affected by *hemin* . Nuclear run-on transcription assays confirmed the increase in the rate of transcription of gamma- and epsilon-globin genes in *hemin* -treated versus untreated hAEC. These results indicate that *hemin* affects the expression of embryonic and fetal globin genes by acting both at the transcriptional and posttranscriptional levels. These results may be relevant to the action of other agents that affect the hemoglobin phenotype of human erythroid cells.

9/3,AB/6
DIALOG(R)File 155:MEDLINE(R)

08736242 96084029 PMID: 8537230
Low oxygen enhances sickle and normal erythropoiesis and *fetal* *hemoglobin* synthesis in vitro.
Weinberg R S; Acosta R; Knobloch M E; Garber M; Alter B P Department of Medicine, Mount Sinai School of Medicine, New York, NY 10029, USA.
Hemoglobin (UNITED STATES) Sep 1995, 19 (5) p263-75, ISSN 0363-0269 Journal Code: 7705865
Contract/Grant No.: HL26132; HL; NHLBI; HL28381; HL; NHLBI Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Erythropoiesis is increased in cultures of human blood progenitors when oxygen tension is reduced from 20% (room air) to 5% (low oxygen, closer to physiological bone marrow levels). The effects of low oxygen on *gamma* -*globin* synthesis and colony growth in methyl cellulose cultures of blood mononuclear cells from normal individuals and patients with sickle cell diseases were examined. Low oxygen increased colony numbers by 1.5- to 2-fold and erythropoietin sensitivity by almost 2-fold. The interval required for maximal colony growth in cultures from patients with sickle cell disease (sickle colonies) was reduced from 17 days in 20% oxygen to 13 days in 5% oxygen. Relative synthesis of *gamma*-*globin* was examined by labeling with 3H-leucine and electrophoresis on Triton acid urea polyacrylamide gels. The % gamma was 1.7-fold higher in normal and 1.4-fold higher in sickle cultures on day 13 in low oxygen. On day 16 the expected temporal decline was not seen in low oxygen, and the % gamma was 2-fold higher in normal and 1.8-fold higher in the sickle studies. *Hemin* increased colony growth and *gamma*-*globin* synthesis in normal cultures in air, and the effects of *hemin* and low oxygen were additive. In sickle cultures, *hemin* and low oxygen had additive effects on colony growth, but only low oxygen increased *gamma*- *globin* synthesis. Interleukin-3 increased colony numbers on day 13,

primarily by acceleration of peak growth. Interleukin-3 also increased *gamma*-*globin* synthesis in low oxygen in normal but not sickle cultures. Thus, low oxygen increases in vitro sensitivity to erythropoietin, colony numbers, and relative *gamma*-*globin* synthesis in normal and sickle cultures.

9/3,AB/7
DIALOG(R)File 155:MEDLINE(R)

08199702 94334299 PMID: 7520036
Differential induction of adult and fetal globin gene expression in the human CML cell subline KU-812F/33.
Endo T; Ishibashi Y; Shiokawa S; Fukumaki Y; Okano H
Department of School-Nurse Training, Kyushu Women's Jr. College, Fukuoka. Journal of biochemistry (JAPAN) Mar 1994, 115 (3) p540-4, ISSN 0021-924X Journal Code: 0376600
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Various chemicals which are known to have positive effects on differentiation of some erythroid cell lines were tested on a human chronic myelogenous leukemia cell line, KU-812F. Succinic acid, 5-azacytidine, daunomycin, and *hemin* showed a positive effect. Among them, *hemin* and 5-azacytidine were the most effective inducers for erythroid differentiation of KU-812F cells. Dimethylsulfoxide, cytosine arabinofuranoside, and sodium n-butyrate showed no effect. In addition, subclone KU-812F/33 derived from the KU-812F cell line showed differential expression of the beta- and *gamma*-*globin* genes in the presence of either 2 microM 5-azacytidine or 40 microM *hemin*. Hemoglobin synthesis in differentiated KU-812F/33 cells was analyzed by isoelectric focusing gel electrophoresis, and S1 mapping analysis of beta- and *gamma*-*globin* mRNA was performed. After treatment with 5-azacytidine, the beta-globin gene expression was predominantly enhanced (18.75-fold higher level of beta-globin mRNA). After treatment with *hemin*, the most notable increase was in the *gamma*-*globin* gene expression (1.83-fold higher level of *gamma*-*globin* mRNA), while no increment of beta-globin was observed.

9/3,AB/8
DIALOG(R)File 155:MEDLINE(R)

07528045 93054960 PMID: 1385454
A novel nuclear protein which binds to G gamma and A *gamma* *globin* promoters and modulates hemoglobin synthesis in K562 cells. Baliga B S; Phillips K; Aliabadi Z; Mankad V N
Molecular Biology Division, University of South Alabama College of Medicine, Mobile 36617.
Journal of cellular biochemistry (UNITED STATES) Aug 1992, 49 (4) p394-8, ISSN 0730-2312 Journal Code: 8205768
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Nuclear extract of human erythroleukemic cell line K562 contains a 70 kDa protein which is gradually reduced when cells are induced to express globin genes by 25 microM *hemin* . When globin synthesis was inhibited by cycloheximide (100 micrograms/ml) or Actinomycin D (1 microgram/ml), the disappearance of this protein was prevented. The 70 kDa nuclear protein exhibited strong binding to G gamma and A *gamma* *globin* promoters but not to beta-globin promoter. This suggests that this 70 kDa nuclear protein may be involved in the regulation of fetal globin gene expression.

9/3,AB/9
DIALOG(R)File 155:MEDLINE(R)

06578747 90275205 PMID: 1693523
Expression of the human *gamma*-*globin* gene after retroviral transfer to transformed erythroid cells.
Rixon M W; Harris E A; Gelinas R E
Program in Molecular Medicine, Fred Hutchinson Cancer Research Center, Seattle, Washington 98104.
Biochemistry (UNITED STATES) May 8 1990, 29 (18) p4393-400, ISSN 0006-2960 Journal Code: 0370623
Contract/Grant No.: DK-31232; DK; NIDDK; HL-36449; HL; NHLBI; HL-37073; HL; NHLBI
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Regulation of the human fetal (*gamma*) *globin* gene and a series of mutant *gamma*-*globin* genes was studied after retroviral transfer into erythroid cells with fetal or adult patterns of endogenous globin gene expression. Steady-state RNA from a virally transferred A *gamma*-*globin* gene with a normal promoter increased after induction of erythroid maturation of murine erythroleukemia cells and comprised from 2% to 23% of the mouse beta maj-globin RNA level. RNA expression from the virally transferred A *gamma*-*globin* gene comprised 23% of the endogenous G gamma- + A *gamma*-*globin* expression in K 562 cells after treatment with *hemin*. Expression from a virally transferred gamma- or beta-globin gene

exceeded endogenous gamma- or beta-globin expression by a factor of 6 or more in the human erythroleukemia line KMOE, in which the endogenous globin genes are weakly inducible. In these experiments, no difference in expression was observed between the gene with the normal promoter and an A *gamma*-*globin* gene with a point mutation in its promoter (-196 C-to-T) that has been associated with hereditary persistence of *fetal* *hemoglobin* (HPFH). To test for cis-acting determinants located within the introns of the *gamma*-*globin* gene, expression was measured from a set of *gamma*-*globin* genes configured with either intron alone or with neither intron. In contrast to an intronless beta-globin gene, which is not expressed in MEL cells, the intronless *gamma*-*globin* gene was expressed in MEL cells at 24% of the level of an intron-containing gene.(ABSTRACT TRUNCATED AT 250 WORDS)

9/3,AB/10
DIALOG(R)File 155:MEDLINE(R)

05868111 88294316 PMID: 3165297
High-resolution analysis of the human *gamma*-*globin* gene promoter in K562 erythroleukemia cell chromatin.
Gimble J M; Max E E; Ley T J
Laboratory of Immunogenetics, NIAID, Bethesda, MD.
Blood (UNITED STATES) Aug 1988, 72 (2) p606-12, ISSN 0006-4971 Journal Code: 7603509
Contract/Grant No.: DK38682; DK; NIDDK
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
We performed high-resolution mapping studies of the DNAse I-hypersensitive sites located just 5' to the human G gamma- and A *gamma*-*globin* genes of K562 erythroleukemia cells, in which these genes are constitutively expressed at low levels. This analysis revealed that the hypersensitive site extends from approximately -210 +/- 5 to -25 +/- 5 base pairs (bp) upstream from the transcription initiation site. Within this region, a GC-rich region located between the proximal CCAAT box and the TATA box is particularly accessible to nuclease digestion; however, the 5' end of the hypersensitive site is less accessible to nucleases. The pattern of DNAse I cleavage does not change on either strand with *hemin* induction of K562 cells, which increases the rate of *gamma*-*globin* gene transcription about threefold. The region within the hypersensitive site includes all the consensus promoter elements of the *gamma*-*globin* genes as well as an octamer sequence located between -182 and -175, and a region associated with a variety of mutations that may cause hereditary persistence of *fetal* *hemoglobin* (HPFH).

9/3,AB/11
DIALOG(R)File 155:MEDLINE(R)

05614550 88037554 PMID: 2444837
Differential induction of adult and fetal globin gene expression in the human erythremia cell line KMOE.
Takihara Y; Rutherford T; Shiokawa S; Fukumaki Y; Endo T; Okano H Department of Biochemistry, Kyushu University, School of Medicine, Fukuoka, Japan.
Leukemia : official journal of the Leukemia Society of America, Leukemia Research Fund, U.K (UNITED STATES) Sep 1987, 1 (9) p673-6, ISSN 0887-6924 Journal Code: 8704895
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Induction of globin gene expression in KMOE cells derived from a patient with acute erythremia was studied by Northern blot and S1 analysis. KMOE cells exposed to cytosine arabinofuranoside (Ara-C) synthesized beta-globin gene transcripts, however, in the presence of *hemin* *gamma*- *globin* gene transcripts. An increase in alpha-globin gene transcripts was also detectable in KMOE cells treated with both Ara-C and *hemin*. Upon exposure to *hemin* after exposure to Ara-C, or exposure to Ara-C after *hemin*, there was a 5-10-fold increase in *gamma*- *globin* gene transcripts compared to that of cells induced by *hemin* alone. Neither epsilon nor zeta globin transcripts were detected. The KMOE cell line, therefore, exhibits phenotypic properties of adult and fetal erythroid cells.

9/3,AB/12
DIALOG(R)File 155:MEDLINE(R)

04819632 85204264 PMID: 2581804
Efficient cell proliferation and predominant accumulation of epsilon-globin mRNA in human leukemic K562 cells which produce mostly Hb Gower 1.
Gambari R; Amelotti F; Piva R
Experientia (SWITZERLAND) May 15 1985, 41 (5) p673-5, ISSN 0014-4754 Journal Code: 0376547
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Long-term cultures of K562(S) cells in 50-75 microM *hemin* allow the selection of '*hemin*-resistant' K562 cells together with cells which proliferate efficiently

while fully induced to express the human embryonic globin genes, as the hemoglobin Gower 1 (zeta 2 epsilon 2) is the predominant hemoglobin produced. Our experiments demonstrate that these K562 cells accumulate mostly epsilon-globin mRNA (epsilon-globin mRNA/ *gamma*-*globin* mRNA = 2.9) suggesting that the control of hemoglobin expression is at a pretranslational level.

9/3,AB/13
DIALOG(R)File 155:MEDLINE(R)

03776823 82047314 PMID: 6945884
Heterogeneity in the cellular commitment of a human leukemic cell line: K 562.
Vainchenker W; Testa U; Guichard J; Titeux M; Breton-Gorius J Blood cells (GERMANY, WEST) 1981, 7 (2) p357-75, ISSN 0340-4684 Journal Code: 7513567
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
The cellular origin of the K 562 cell line, established from a patient in the blast crisis of chronic myeloid leukemia has been investigated. In agreement with previous reports, an erythroid differentiation was observed. A minority of immature, but hemoglobinized erythroblasts were identified both by electron microscopy and by immunofluorescence using an antibody to *gamma*-*globin* chains. Embryonic and *fetal* *hemoglobin* (Hb) were synthesized. *Hemin* increased the number of erythroblasts as well as the absolute amount of Hb synthesized: the Hb pattern was also significantly modified. By cytochemical ultrastructural detection of peroxidase activity (PA), a weak PA, distinct from granulocytic peroxidases, was found exclusively in the nuclear envelope and rough endoplasmic reticulum in a small number proportion of cells. In its localization this PA resembled that of normal and leukemic promegakaryoblasts. The addition of sodium butyrate or dimethylformamide markedly increased the number of these cells (up to 30%) but did not modify their cytoplasmic maturation. No modification of Hb synthesis was observed. Cloning of the K 562 line revealed a marked heterogeneity from one clone to another in Hb production, in the phenotype of Hb synthesis, and in the inducibility by butyrate or dimethylformamide. An inverse relationship between the number of cells with PA and Hb production was found in the different clones. Recloning some of these primary clones resulted in secondary clones, which displayed properties similar to those from which they had originated. All attempts to obtain granulocytic differentiation by addition of different inducers failed. These results clearly indicate that the K 562 cell line arises from the proliferation of bipotent stem cells, these cells possessing variable capacities of differentiation toward erythroid and presumably megakaryocytic cell lineages.
? ds

Set	Items	Description
S1	54	GAMMA()GLOBIN AND HEMIN
S2	21	S1 AND INDUCTION
S3	2	S2 AND ASSAY
S4	3977	FETAL()HEMOGLOBIN
S5	41	S4 AND HEMIN
S6	4	S5 AND ASSAY
S7	2	S6 NOT S3
S8	15	S1 AND S4
S9	13	S8 NOT S6

? s s4 and induction not s8
3977 S4
222499 INDUCTION
15 S8
S10 85 S4 AND INDUCTION NOT S8
? s s10 and induc?(3a)(gamma or fegal)
85 S10
0 INDUC?(3A)(GAMMA
0 FEGAL)
S11 0 S10 AND INDUC?(3A)(GAMMA OR FEGAL)
? s s10 and induc?(3n)(gamma or fetgal)
85 S10
1352472 INDUC?
188915 GAMMA
0 FETGAL
11434 INDUC?(3N)(GAMMA OR FETGAL)
S12 14 S10 AND INDUC?(3N)(GAMMA OR FETGAL)
? s s10 and induc?(3n)(gamma or fetal)
85 S10
1352472 INDUC?
188915 GAMMA
193735 FETAL
18002 INDUC?(3N)(GAMMA OR FETAL)
S13 50 S10 AND INDUC?(3N)(GAMMA OR FETAL)
? t s13/3,ab/all

13/3,AB/1
DIALOG(R)File 155:MEDLINE(R)

14175782 22370403 PMID: 12482403
Induction of *Fetal* *Hemoglobin* by Propionic and Butyric Acid Derivatives: Correlations between Chemical Structure and Potency of Hb F *Induction*.
Liakopoulou Effie; Li Qiliang; Stamatoyannopoulos George; et al Blood cells, molecules & diseases (United States) Jul-Aug 2002, 29 (1) p48-56, ISSN 1079-9796 Journal Code: 9509932

Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: In Process
ABSTRACTShort-chain fatty acids (C2-C9) *induce* *fetal* *hemoglobin* synthesis in primary cell cultures, primates, and patients. We carried out experiments to test whether relationships exist between chemical structure and the Hb F-inducing potential of several short-chain fatty acid derivatives. BFUe cultures were performed in the presence of propionic and butyric congeners, covering the full spectrum of substitutions of these molecules, including polar and non-polar groups, esters, and double bonds. We found that the *fetal* *hemoglobin* *inducibility* is related to the chemical structure of the inducing compound. This structure-activity relation depends on the length of carbon chain, the nature of the substitutions, and the position of more potent substitutions on the carbon chain. It appears that substitutions enhancing the inducibility of these compounds are (with decreasing potency): methyl > phenyl > hydroxy >> amino groups. Placement of these substitutions at a position distal to the carboxyl group enhances *gamma*-globin *inducibility*. Presence of the carboxyl group is prerequisite for *gamma*-globin *inducibility*.

13/3,AB/2
DIALOG(R)File 155:MEDLINE(R)

14119548 22356402 PMID: 12468915
Induction of *fetal* *hemoglobin* synthesis in children with sickle cell anemia on low-dose oral sodium phenylbutyrate therapy. Resar Linda M S; Segal Jodi B; Fitzpatric Lorna K; Friedmann Alison; Brusilow Saul W; Dover George J; et al
Journal of pediatric hematology/oncology - official journal of the American Society of Pediatric Hematology/Oncology (United States) Dec 2002, 24 (9) p737-41, ISSN 1077-4114 Journal Code: 9505928
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: In Process
This study was designed to determine if low doses of oral sodium phenylbutyrate (SPB) induce hemoglobin F (HbF) synthesis in children with hemoglobin SS (HbSS). We treated 8 children with HbSS over a period of 5-30 weeks. The initial dose (1.0 g/d) was increased weekly (by 1.0 g/d) until F-reticulocytes doubled. All patients showed an increase in F-reticulocytes (= 0.002) that was dose-dependent (= 0.001). Three of 5 patients who continued oral SPB for more than 10 weeks had substantial increases in HbF. We conclude that lower dose SPB is effective in inducing HbF synthesis in some children with HbSS. Further trials are warranted to determine the optimal treatment regimen.

13/3,AB/3
DIALOG(R)File 155:MEDLINE(R)

13699852 22208885 PMID: 12221674
Induction of *fetal* *hemoglobin* synthesis by valproate: modulation of MAP kinase pathways.
Witt O; Monkemeyer S; Kanbach K; Pekrun A
Laboratory for Hematological and Cancer Research, Children's Hospital, University of Gottingen, Gottingen, Germany. owitt@gwdg.de American journal of hematology (United States) Sep 2002, 71 (1) p45-6, ISSN 0361-8609 Journal Code: 7610369
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Valproate has been found to stimulate *fetal* *hemoglobin* (HbF) synthesis in patients with sickle cell disease. In accordance with these clinical observations, we found a moderate *induction* of HbF synthesis in K562 erythroid cells in vitro. Investigation of the role of the mitogen-activated protein kinase (MAPK) pathways by Western blot analysis and use of specific kinase inhibitors suggests that inhibition of ERK pathway and activation of the p38 pathway may contribute to the HbF-inducing activity of valproate. Copyright 2002 Wiley-Liss, Inc.

13/3,AB/4
DIALOG(R)File 155:MEDLINE(R)

12818652 21491122 PMID: 11605172
Pharmacological *induction* of *fetal* *hemoglobin* in sickle cell disease and beta-thalassemia.
Atweh G F; Loukopoulos D
Division of Hematology, Mount Sinai School of Medicine, New York, NY 10029, USA.
Seminars in hematology (United States) Oct 2001, 38 (4) p367-73, ISSN 0037-1963 Journal Code: 0404514
Document type: Journal Article; Review; Review, Tutorial
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
A number of pharmacological agents are currently available for the *induction* of *fetal* *hemoglobin* (HbF) in patients with sickle cell disease and beta-thalassemia. Here we review the development of this new class of therapeutics and summarize the clinical trials that investigate their efficacy in patients with hemoglobin disorders. Hydroxyurea is the first of these drugs to be approved by the Food and Drug Administration for the treatment of sickle cell disease.

Currently, the major focus is the development of safer agents and combinations of drugs that can increase HbF to levels high enough to prevent all complications of the disease. Progress in adapting the same strategy to the treatment of thalassemic disorders has been much slower. Although all the agents that are effective in sickle cell disease have similar HbF-inducing activity in beta-thalassemia, their use has rarely resulted in significant amelioration of the anemia. More research and more effective agents will be needed to make a significant impact on thalassemia. Nonetheless, success in this relatively young field has been very gratifying; before the end of this decade, clinically meaningful *induction* of HbF may become an achievable goal in most patients with hemoglobin disorders. Copyright 2001 by W.B. Saunders Company.

13/3,AB/5
DIALOG(R)File 155:MEDLINE(R)

12818648 21491118 PMID: 11605168
Nitric oxide therapy in sickle cell disease.
Gladwin M T; Schechter A N
Critical Care Medicine Department, Warren G. Magnuson Clinical Center, NIH, Bethesda, MD 20892-1662, USA.
Seminars in hematology (United States) Oct 2001, 38 (4) p333-42, ISSN 0037-1963 Journal Code: 0404514
Document type: Journal Article; Review; Review, Tutorial
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Recent clinical and experimental data suggest that nitric oxide (NO) may play a role in the pathogenesis and therapy of sickle cell disease. NO, a soluble gas continuously synthesized in endothelial cells by the NO synthase (NOS) enzyme systems, regulates basal vascular tone and endothelial function, and maintains blood oxygenation via hypoxic pulmonary vasoconstriction and reduced shunt physiology. These vital homeostatic processes may be impaired in sickle cell disease and contribute to its pathogenesis. Therapeutic NO inhalation exerts significant direct effects on the pulmonary vasculature to reduce pulmonary pressures and increase oxygenation that may prove beneficial in acute chest syndrome and secondary pulmonary hypertension. Delivery of NO bound to hemoglobin or in plasma may improve blood flow and hemoglobin saturation, and thus reduce ischemia-reperfusion injury. Other NO-related effects on adhesion molecule expression and *fetal* *hemoglobin* *induction* are of interest. While direct evidence for a clinical benefit of NO therapy in sickle cell disease has not been reported, studies are underway to determine if inhaled NO will reduce the substantial morbidity and mortality suffered by these patients.

13/3,AB/6
DIALOG(R)File 155:MEDLINE(R)

11296929 21335942 PMID: 11442489
Accumulation of *gamma*-globin mRNA and *induction* of erythroid differentiation after treatment of human leukaemic K562 cells with tallimustine.
Bianchi N; Chiarabelli C; Borgatti M; Mischiati C; Fibach E; Gambari R Department of Biochemistry and Molecular Biology, University of Ferrara, Italy.
British journal of haematology (England) Jun 2001, 113 (4) p951-61, ISSN 0007-1048 Journal Code: 0372544
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Human leukaemic K562 cells can be induced in vitro to erythroid differentiation by a variety of chemical compounds, including haemin, butyric acid, 5-azacytidine, cytosine arabinoside, mithramycin and chromomycin, cisplatin and cisplatin analogues. Differentiation of K562 cells is associated with an increase of expression of embryo-fetal globin genes, such as the zeta-, epsilon- and gamma-globin genes. The K562 cell line has been proposed as a very useful in vitro model system to determine the therapeutic potential of new differentiating compounds as well as to study the molecular mechanism(s) regulating changes in the expression of embryonic and *fetal* human globin genes. *Inducers* of erythroid differentiation stimulating gamma-globin synthesis could be considered for possible use in the therapy of haematological diseases associated with a failure in the expression of normal beta-globin genes. We have analysed the effects of tallimustine and distamycin on cell growth and differentiation of K562 cells. The results demonstrated that tallimustine is a potent inducer, while distamycin is a weak inducer, of K562 cell erythroid differentiation. Erythroid differentiation was associated with an increase of accumulation of gamma-globin mRNA and of production of both haemoglobin (Hb) Gower 1 and Hb Portland. In addition, tallimustine-mediated erythroid *induction* occurred in the presence of activation of the apoptotic pathway. The reasons for proposing tallimustine as an *inducer* of *gamma* -globin gene expression are strongly sustained by the finding that this compound stimulates fetal haemoglobin production in human erythroid precursor cells from normal subjects.

13/3,AB/7
DIALOG(R)File 155:MEDLINE(R)

11220706 21257628 PMID: 11358353

In vivo silencing of the human gamma-globin gene in murine erythroid cells following retroviral transduction.

Lung H Y; Meeus I S; Weinberg R S; Atweh G F

Department of Medicine, Mount Sinai School of Medicine, One Gustave L. Levy Place, New York, NY 10029, USA.

Blood cells, molecules & diseases (United States) Dec 2000, 26 (6) p613-9, ISSN 1079-9796 Journal Code: 9509932

Contract/Grant No.: HL28381; HL; NHLBI; HL54184; HL; NHLBI Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Increased expression of *fetal* *hemoglobin* can ameliorate the clinical severity of sickle cell disease. Whereas temporary *induction* of *fetal* *hemoglobin* can be achieved by pharmacologic therapy, gene transfer resulting in high-level expression of the fetal gamma-globin gene may provide a permanent cure for sickle cell disease. We had previously developed a high-titer, genetically stable retroviral vector in which the human gamma-globin gene was linked to HS-40, the major regulatory element of the human alpha-globin gene cluster. Based on experience in transgenic mice, the truncated promoter of the gamma-globin gene of this vector should be active in adult erythroid cells. Our earlier studies demonstrated that this retroviral vector can give rise to high-level expression of the human gamma-globin gene in murine erythroleukemia (MEL) cells. We have now utilized this vector to transduce murine bone marrow cells that were transplanted into W/W(v) recipient mice. Analysis of transduction of murine BFU-e's in vitro and peripheral blood cells from transplanted mice in vivo demonstrated efficient transfer of the human gamma-globin gene. However, in contrast to the high level of expression of the human gamma-globin gene of this vector in MEL cells, the gene was completely silent in vivo in all transplanted mice. These observations confirm that all the necessary regulatory elements responsible for the developmental stage-specific expression of the human gamma-globin gene reside in its proximal sequences. They also emphasize the differences between gene regulation in MEL cells, transgenic mice, and retroviral gene transfer vectors. For this form of globin gene therapy to succeed, the proximal regulatory elements of the human gamma-globin gene may have to be replaced with different regulatory elements that allow the expression of the gamma-globin coding sequences in adult red cells in vivo. Copyright 2000 Academic Press.

13/3,AB/8
DIALOG(R)File 155:MEDLINE(R)

11216893 21240433 PMID: 11342457

Short-chain fatty acid derivatives stimulate cell proliferation and induce STAT-5 activation.

Boosalis M S; Bandyopadhyay R; Bresnick E H; Pace B S; Van DeMark K; Zhang B; Faller D V; Perrine S P

Department of Medicine, Cancer Research Center and Hemoglobinopathy-Thalassemia Research Unit, Boston University School of Medicine, Boston, MA, USA.

Blood (United States) May 15 2001, 97 (10) p3259-67, ISSN 0006-4971 Journal Code: 7603509

Contract/Grant No.: CA-084193; CA; NCI; DK-52962; DK; NIDDK; HL-61208; HL ; NHLBI

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Current chemotherapeutic and butyrate therapeutics that *induce* *fetal* *hemoglobin* expression generally also suppress erythropoiesis, limiting the production of cells containing *fetal* *hemoglobin* (F cells). Recently, selected short-chain fatty acid derivatives (SCFADs) were identified that *induce* endogenous *gamma* -globin expression in K562 cells and human burst-forming units-erythroid and that increase proliferation of human erythroid progenitors and a multilineage interleukin-3-dependent hematopoietic cell line. In this report, *gamma*-globin *inducibility* by these SCFADs was further demonstrated in mice transgenic for the locus control region and the entire beta-globin gene locus in a yeast artificial chromosome and in 2 globin promoter-reporter assays. Conditioned media experiments strongly suggest that their proliferative activity is a direct effect of the test compounds. Investigation of potential mechanisms of action of these SCFADs demonstrates that these compounds induce prolonged expression of the growth-promoting genes c-myb and c-myc. Both butyrate and specific growth-stimulatory SCFADs induced prolonged signal transducer and activator of transcription (STAT)-5 phosphorylation and activation, and c-cis expression, persisting for more than 120 minutes, whereas with IL-3 alone phosphorylation disappeared within minutes. In contrast to butyrate treatment, the growth-stimulating SCFADs did not result in bulk histone H4 hyperacetylation or *induction* of p21(Waf/Cip), which mediates the suppression of cellular growth by butyrate. These findings suggest that the absence of bulk histone hyperacetylation and p21 *induction*, but prolonged *induction* of cis, myb, myc, and STAT-5 activation, contribute to the cellular proliferation induced by selected SCFADs.

13/3,AB/9
DIALOG(R)File 155:MEDLINE(R)

11121330 21127594 PMID: 11224687
Pharmacologic *induction* of *fetal* *hemoglobin*: raising the therapeutic bar in sickle cell disease.
Atweh G F; Schechter A N
Division of Hematology, Department of Medicine, Mount Sinai School of Medicine, New York, New York 10029, USA. george.atweh@mssm.edu Current opinion in hematology (United States) Mar 2001, 8 (2) p123-30, ISSN 1065-6251 Journal Code: 9430802
Document type: Journal Article; Review; Review, Tutorial
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
The favorable effects of high levels of *fetal* *hemoglobin* (Hb F) in sickle cell disease have been recognized for several decades. This has been an important incentive for the development of therapeutic agents that increase Hb F production. 5-Azacytidine, the first such agent in clinical use, was proposed based on a molecular understanding of the role of DNA methylation in globin gene regulation. Controversy over the mechanism of Hb F *induction* by 5-azacytidine led to the identification of hydroxyurea as another agent that can increase Hb F production. Although the clinical benefit of hydroxyurea has been demonstrated in a randomized clinical trial, greater increases in Hb F are clearly needed for optimal therapeutic effect. Butyrates also increase Hb F levels, and their use in combination with hydroxyurea appears to be synergistic. Now that multiple therapeutic agents are available for Hb F *induction* , the use of combination therapy to increase Hb F levels sufficiently to prevent all the complications of sickle cell disease has become a realistic goal.

13/3,AB/10
DIALOG(R)File 155:MEDLINE(R)

11089463 21104342 PMID: 11170226
Hydroxyurea-induced oxidative damage of normal and sickle cell hemoglobins in vitro: amelioration by radical scavengers.
Iyamu E W; Fasold H; Roa D; del Pilar Aguinaga M; Asakura T; Turner E A Comprehensive Sickle Cell Center, Meharry Medical College, Nashville, Tennessee, USA.
Journal of clinical laboratory analysis (United States) 2001, 15 (1) p1-7, ISSN 0887-8013 Journal Code: 8801384
Contract/Grant No.: 2P60-HL33737; HL; NHLBI; 2P60-HL38632; HL; NHLBI; U24-HL 58930; HL; NHLBI
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Hydroxyurea (HU) *induces* *fetal* *hemoglobin* (Hb F) production in patients with sickle cell anemia. The therapeutic dosage of HU used for Hb F *induction* often elicits myelosuppression, which becomes its major associated complication. We examined the effect of HU on hemoglobin modulation and the role of radical scavengers on these induced changes. In vitro exposure of human blood to various concentrations of HU at predetermined time intervals induced a progressive dose-dependent oxidation (MetHb formation) of both adult (Hb AA) and sickle (Hb SS) hemoglobins. The oxidative effect of HU on Hb SS was 3 times greater than its effect on Hb AA. Similar but less profound changes were observed in H2O2-treated samples. Hb F was, however, observed to be relatively resistant to HU-induced oxidative damage. A substantial protective effect of Hb by alpha-tocopherol, ascorbic acid, and D-mannitol was observed during pretreatment of Hb AA and Hb SS blood samples. Analyses of the hemoglobins and their globin chain components by high-performance liquid chromatography revealed a considerable protective effect by these free radical scavengers. These results indicate that the HU-induced damage of hemoglobin and their component globin chains can be reduced by radical scavengers.

13/3,AB/11
DIALOG(R)File 155:MEDLINE(R)

10953265 20529834 PMID: 11074540
Mechanism for *fetal* *hemoglobin* *induction* by hydroxyurea in sickle cell erythroid progenitors.
Baliga B S; Pace B S; Chen H H; Shah A K; Yang Y M
Department of Pediatrics and Comprehensive Sickle Cell Center, University of South Alabama College of Medicine, Mobile 36617, USA.
American journal of hematology (UNITED STATES) Nov 2000, 65 (3) p227-33, ISSN 0361-8609 Journal Code: 7610369
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Hydroxyurea (HU) is a widely used cytotoxic agent that is known to *induce* *fetal* *hemoglobin* (HbF) production and is presently used to ameliorate the severity of pain episodes in patients with sickle cell anemia (HbSS). Previously we have shown that HU inhibits growth of burst forming unit-erythroid (BFU-E) colonies in a dose-dependent manner, while *fetal* *hemoglobin* levels were increased. In the present report, we extended our analysis demonstrating the number of S phase cells is significantly higher for HbSS patients that respond to HU therapy. Studies were completed in vitro using erythroid progenitors

derived from umbilical cord samples or peripheral blood from patients with HbS-hereditary persistence of *fetal* *hemoglobin* (HbS-HPFH) or HbSS disease. The effect of HU on (a) S phase erythroid progenitors, (b) BFU-E colony growth, (c) HbF levels in BFU-E colonies, and (d) total cellular RNA synthesis was analyzed in vitro for the three groups. The level of S phase erythroid progenitors was similar for all three groups and BFU-E colony growth was inhibited 92-94% for all samples in a dose-dependent manner. The HbF levels were increased in BFU-E colonies from HbSS patients (control, 4.0% +/- 1.15% vs. +HU, 22.67% +/- 2.03%) whereas HbF levels were decreased in BFU-E colonies derived from umbilical cord samples (control, 80% +/- 9.07% vs. +HU, 35.7% +/- 4.81%) or HbS-HPFH patients (control, 49.67% +/- 3.84% vs. +HU, 23.3% +/- 0.88%). Total RNA synthesis measured by 3H-uridine incorporation increased with increasing concentrations of HU; however, actinomycin D inhibited HU-induced RNA synthesis. These results suggest that HU can inhibit an active globin gene without preference and that newly synthesized RNA is under transcriptional control mechanisms.

13/3,AB/12
DIALOG(R)File 155:MEDLINE(R)

10901193 20458779 PMID: 11001887
2-deoxy 5-azacytidine and *fetal* *hemoglobin* *induction* in sickle cell anemia.
Koshy M; Dorn L; Bressler L; Molokie R; Lavelle D; Talischy N; Hoffman R; van Overveld W; DeSimone J
University of Illinois at Chicago, Chicago, IL, USA. mkoshy@uic.edu Blood (UNITED STATES) Oct 1 2000, 96 (7) p2379-84, ISSN 0006-4971 Journal Code: 7603509
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Augmentation of the *fetal* *hemoglobin* (HbF) levels is of therapeutic benefit in patients with sickle cell anemia. Hydroxyurea (HU), by increasing HbF, lowers rates of pain crisis, episodes of acute chest syndrome, and requirements for blood transfusions. For patients with no HbF elevation after HU treatment, augmentation of HbF levels by 5-aza-2'-deoxycytidine (5-aza-CdR, decitabine) could serve as an alternate mode of treatment. Eight adult patients participated in a dose-escalating phase I/II study with 5-aza-CdR at doses ranging from 0.15 to 0.30 mg/kg given 5 days a week for 2 weeks. HbF, F cell, F/F cell, gamma-globin synthesis ratio, complete blood count, and chemistry were measured. The average gamma-globin synthesis relative to non-alpha-globin synthesis prior to therapy was 3.19% +/- 1.43% and increased to 13.66% +/- 4.35% after treatment. HbF increased from 3.55% +/- 2.47% to 13.45% +/- 3.69%. F cells increased from 21% +/- 14.8% to 55% +/- 13.5% and HbF/F cell increased from 17% to 24%. In the HU nonresponders HbF levels increased from 2.28% +/- 1.61% to 2.6% +/- 2.15% on HU, whereas on 5-aza-CdR HbF increased to 12.70% +/- 1.81%. Total hemoglobin increased by 1 g/dL in 6 of 8 patients with only minor reversible toxicities, and all patients tolerated the drug. Maximum HbF was attained within 4 weeks of treatment and persisted for 2 weeks before falling below 90% of the maximum. Therefore 5-aza-CdR could be effective in increasing HbF in patients with sickle cell anemia who failed to increase HbF with HU. Demonstration of sustained F levels with additional treatment cycles without toxicity is currently being performed.

13/3,AB/13
DIALOG(R)File 155:MEDLINE(R)

10692677 20243302 PMID: 10779423
Improvement of erythropoiesis in beta-thalassemic mice by continuous erythropoietin delivery from muscle.
Bohl D; Bosch A; Cardona A; Salvetti A; Heard J M
Laboratoire Retrovirus et Transfert Genetique, Institut Pasteur, Paris, France.
Blood (UNITED STATES) May 1 2000, 95 (9) p2793-8, ISSN 0006-4971 Journal Code: 7603509
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
beta-Thalassemias are highly prevalent genetic disorders that can cause severe hemolytic anemia. The main pathophysiologic feature of beta-thalassemia is the accumulation of unpaired alpha-globin chains in erythrocyte precursors and red blood cells (RBCs). This accumulation alters cell membrane function and results in early cell destruction and ineffective erythropoiesis. Correction of globin chain imbalance through the *induction* of *fetal* *hemoglobin* (HbF) synthesis is a tentative therapeutic approach for this class of diseases. In short-term in vitro or in vivo assays, recombinant human erythropoietin increases the frequency of erythroid precursors programmed to HbF in humans and to beta-minor globin in mice. In contrast, long-term treatment of beta-thalassemic patients did not induce HbF significantly. We took advantage of highly efficient adeno-associated virus-mediated (AAV-mediated) gene transfer into mouse muscle to induce a robust and sustained secretion of mouse erythropoietin in beta-thalassemic mice, which represent a suitable model for human beta-thalassemia intermedia.

A 1-year follow-up of 12 treated animals showed a stable correction of anemia associated with improved RBC morphology, increased beta-minor globin synthesis, and decreased amounts of alpha-globin chains bound to erythrocyte membranes. More effective erythropoiesis probably accounted for a reduction of erythroid cell proliferation, as shown by decreased proportions of circulating reticulocytes and by reduced iron 59 ((59)Fe) incorporation into erythroid tissues. This study indicates that the continuous delivery of high amounts of autologous erythropoietin induced a sustained stimulation of beta-minor globin synthesis and a stable improvement of erythropoiesis in the beta-thalassemic mouse model. (Blood. 2000;95:2793-2798)

13/3,AB/14
DIALOG(R)File 155:MEDLINE(R)

10514350 20042805 PMID: 10575551
A combination of hydroxyurea and isobutyramide to *induce* *fetal* *hemoglobin* in transgenic mice is more hematotoxic than the individual agents.
Buller A M; Elford H L; DuBois C C; Meyer J; Lloyd J A
Department of Human Genetics, Medical College of Virginia, Virginia Commonwealth University, Richmond 23298-0033, USA.
Blood cells, molecules & diseases (UNITED STATES) Jun-Aug 1999, 25 (3-4) p255-69, ISSN 1079-9796 Journal Code: 9509932
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Pharmacologic agents such as hydroxyurea (HU), N, 3-4 trihydroxybenzamide (didox), and isobutyramide (ISB) can elevate gamma-globin as a potential treatment for the beta-hemoglobinopathies. In these experiments, transgenic mice with 5'HS2 from the human beta-globin locus control region, the fetal (A gamma), and adult (beta s) globin genes were used. Mice were treated with HU, didox, or ISB individually, or with combinations of HU or didox with ISB. The aim was to determine whether these drugs have synergistic effects on the *induction* of *fetal* *hemoglobin* (HbF) and whether the combination regimens are more hematotoxic. In the combination regimens, injections of HU or didox for five weeks were concomitant with ISB treatment every other day for the final three weeks of treatment. The combination of HU + ISB was more hematotoxic than the individual drugs based on significantly increased percentages of reticulocytes and reduced hemoglobin, indicating that caution should be taken in treatments involving combinations of these types of drugs. The didox + ISB combination was not more hematotoxic than the individual drugs. HbF was not induced in the groups treated with the combinations of HU or didox with ISB compared to the individual agents. There was a negligible effect on the percentage of HbF and an unexpected negative effect on the percentage of F cells. The results also have implications for future testing of HbF-inducing drugs in mouse models. In control mice that were phlebotomized but not treated with any drugs, increased percentages of F cells were observed, indicating that blood sampling can cause this effect. In addition, increases in the percentage of F cells did not correlate with increases in the percentage of HbF, indicating that monitoring F cells alone is not a sufficient measure of HbF *induction*.

13/3,AB/15
DIALOG(R)File 155:MEDLINE(R)

10327029 99303843 PMID: 10373600
Peptide nucleic acid (PNA) binding-mediated *induction* of human *gamma*-globin gene expression.
Wang G; Xu X; Pace B; Dean D A; Glazer P M; Chan P; Goodman S R; Shokolenko I
Department of Structural and Cellular Biology, University of South Alabama College of Medicine, MSB 2042, Mobile, AL 36688-0002, USA. gwang@jaguar1.usouthal.edu
Nucleic acids research (ENGLAND) Jul 1 1999, 27 (13) p2806-13, ISSN 0305-1048 Journal Code: 0411011
Contract/Grant No.: HL38639-10; HL; NHLBI
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Peptide nucleic acids (PNAs) can bind to homopurine/homopyrimidine sequences of double-stranded DNA targets in a sequence-specific manner and form [PNA]2/DNA triplexes with single-stranded DNA D-loop structures at the PNA binding sites. These D-loop structures have been found to have a capacity to initiate transcription in vitro. If this strategy can be used to induce transcription of endogenous genes, it may provide a novel approach for gene therapy of many human diseases. Human [beta] globin disorders such as sickle cell anemia and beta-thalassemia are very common genetic diseases that are caused by mutations in the beta-globin gene. When gamma-globin genes are highly expressed in sickle cell patients, the presence of high levels of *fetal* *hemoglobin* (HbF, alpha2gamma2) can compensate for the defective beta-globin gene product and such patients have much improved symptoms or are free of disease. However, the gamma-globin genes are developmentally regulated and normally expressed at very low levels (>1%) in adult blood cells. We have

investigated the possibility of *inducing* *gamma*-globin gene expression with PNAs. Using PNAs designed to bind to the 5' flanking region of the *gamma* -globin gene, *induction* of expression of a reporter gene construct was demonstrated both in vitro and in vivo. More importantly, PNA-mediated *induction* of endogenous *gamma*-globin gene expression was also demonstrated in K562 human erythroleukemia cells. This result suggests that *induction* of *gamma* -globin gene expression with PNAs might provide a new approach for the treatment of sickle cell disease. PNA-induced gene expression strategy also may have implications in gene therapy of other diseases such as genetic diseases, cancer and infectious diseases.

13/3,AB/16
DIALOG(R)File 155:MEDLINE(R)

10185580 99168964 PMID: 10068649
Sustained *induction* of *fetal* *hemoglobin* by pulse butyrate therapy in sickle cell disease.
Atweh G F; Sutton M; Nassif I; Boosalis V; Dover G J; Wallenstein S; Wright E; McMahon L; Stamatoyannopoulos G; Faller D V; Perrine S P
Departments of Medicine, Pediatrics and Biomathematical Sciences, Mount Sinai School of Medicine, New York, NY, USA.
Blood (UNITED STATES) Mar 15 1999, 93 (6) p1790-7, ISSN 0006-4971 Journal Code: 7603509
Contract/Grant No.: HL-15157; HL; NHLBI; HL-37119; HL; NHLBI; HL-54184; HL; NHLBI; +
Comment in Blood. 1999 Mar 15;93(6) 1787-9; Comment in PMID 10068648 Document type: Clinical Trial; Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
High levels of *fetal* *hemoglobin* (Hb F) protect from many of the complications of sickle cell disease and lead to improved survival. Butyrate and other short chain fatty acids were previously shown to increase Hb F production in erythroid cells in vitro and in animal models in vivo. However, butyrates are also known to inhibit the proliferation of many cell types, including erythroid cells. Experience with the use of butyrate in animal models and in early clinical trials demonstrated that the Hb F response may be lost after prolonged administration of high doses of butyrate. We hypothesized that this loss of response may be a result of the antiproliferative effects of butyrate. We designed a regimen consisting of intermittent or pulse therapy in which butyrate was administered for 4 days followed by 10 to 24 days with no drug exposure. This pulse regimen *induced* *fetal* globin gene expression in 9 of 11 patients. The mean Hb F in this group increased from 7.2% to 21.0% (P <.002) after intermittent butyrate therapy for a mean duration of 29.9 weeks. This was associated with a parallel increase in the number of F cells and F reticulocytes. The total hemoglobin levels also increased from a mean of 7.8 g/dL to a mean of 8.8 g/dL (P <.006). The increased levels of Hb F were sustained in all responders, including 1 patient who has been on pulse butyrate therapy for more than 28 months. This regimen, which resulted in a marked and sustained increase in Hb F levels in more than two thirds of the adult sickle cell patients enrolled in this study, was well tolerated without adverse side effects. These encouraging results require confirmation along with an appropriate evaluation of clinical outcomes in a larger number of patients with sickle cell disease.

13/3,AB/17
DIALOG(R)File 155:MEDLINE(R)

10185579 99168963 PMID: 10068648
Induction of *fetal* *hemoglobin* in sickle cell disease. Bunn H F
Division of Hematology, Brigham and Women's Hospital, Boston, MA, USA. Blood (UNITED STATES) Mar 15 1999, 93 (6) p1787-9, ISSN 0006-4971 Journal Code: 7603509
Comment on Blood. 1999 Mar 15;93(6) 1790-7; Comment on PMID 10068649 Document type: Comment; Journal Article; Review; Review, Tutorial Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

13/3,AB/18
DIALOG(R)File 155:MEDLINE(R)

10175351 99160202 PMID: 10052837
Induction of *fetal* *hemoglobin* synthesis with recombinant human erythropoietin in anemic patients with heterozygous beta-thalassemia during pregnancy.
Breymann C; Fibach E; Visca E; Huettner C; Huch A; Huch R Department of Gynaecology and Obstetrics, Clinic of Obstetrics, University of Zurich, Switzerland. chb@fhk.usz.ch
Journal of maternal-fetal medicine (UNITED STATES) Jan-Feb 1999, 8 (1) p1-7, ISSN 1057-0802 Journal Code: 9211288
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
OBJECTIVE: Recombinant human erythropoietin (rhEPO) increases *fetal* *hemoglobin* synthesis in nonpregnant thalassaemic patients. We used rhEPO in 4

pregnant patients with heterozygous beta-thalassemia and anemia to study its effect on erythropoiesis, F cell production, and HbF synthesis. METHODS: Patients were treated with a combination therapy of rhEPO and iron. The effect on HbF synthesis was assessed by the percentage of F reticulocytes, F cells, and total HbF, erythropietis by reticulocyte count, and hemoglobin measurements and iron status by ferritin levels, transferrin saturation, and percentage of hypochromic red cells. RESULTS: RhEPO caused an increase of F reticulocytes (1.5 to 10.5 fold), F cells (5.0 to 7.7 fold), and HbF (1.4 to 2.2 fold). All patients showed an increase of young, immature reticulocytes and had elevated reticulocytes at the end of therapy. Hemoglobin increased with a range from 0.3 to 1.5 g/dL. Transferrin saturation and ferritin levels were normal at the end of the study. There was an increase of the percentage of hypochromic red cells, indicating functional iron deficiency after rhEPO administration despite supplemental iron. CONCLUSIONS: RhEPO stimulates both HbF synthesis and erythropoiesis in pregnant patients with heterozygous beta-thalassemia and anemia. Since it is known that high HbF levels ameliorate thalassemia symptoms in nonpregnant patients, use of rhEPO for the treatment of severe anemia in thalassaemic patients during pregnancy might be further evaluated.

13/3,AB/19
DIALOG(R)File 155:MEDLINE(R)

09936481 98358762 PMID: 9691002
Fetal gene reactivation.
Swank R A; Stamatoyannopoulos G
Division of Medical Genetics, University of Washington, Seattle 98195-7720, USA. raswank@u.washington.edu
Current opinion in genetics & development (ENGLAND) Jun 1998, 8 (3) p366-70, ISSN 0959-437X Journal Code: 9111375
Document type: Journal Article; Review; Review, Tutorial
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Reactivation of silent fetal or embryonic genes could be used for the treatment of genetic diseases caused by mutations of genes normally expressed during the adult stage of development. A paradigm of this approach is the activation of *fetal* *hemoglobin* synthesis in adult individuals and its use in the treatment of beta chain hemoglobinopathies. The current understanding of the molecular control of the beta globin locus is reviewed, as are the cellular and molecular basis of *induction* of *fetal* *hemoglobin* in the adult and the approaches used for stimulation of *fetal* *hemoglobin* synthesis in patients with beta chain hemoglobinopathies.

13/3,AB/20
DIALOG(R)File 155:MEDLINE(R)

09893953 98333129 PMID: 9668532
Elimination of transfusions through *induction* of *fetal* *hemoglobin* synthesis in Cooley's anemia.
Olivieri N F; Rees D C; Ginder G D; Thein S L; Waye J S; Chang L; Brittenham G M; Weatherall D J
Hemoglobinopathy Program, Hospital for Sick Children, Toronto, Ontario, Canada.
Annals of the New York Academy of Sciences (UNITED STATES) Jun 30 1998, 850 p100-9, ISSN 0077-8923 Journal Code: 7506858
Contract/Grant No.: DDK 29902; DK; NIDDK
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Pharmacological stimulation of *fetal* *hemoglobin* production is of considerable interest as an alternative approach to therapy for Cooley's anemia. While intravenous compounds have been effective in inducing short-term increases in *fetal* *hemoglobin* in a few patients, long-term elimination of transfusion requirement has not been reported. In patients with Cooley's anemia, treatment with oral sodium phenylbutyrate alone, sodium phenylbytyrate combined with hydroxyurea, and hydroxyurea alone, has augmented *fetal* *hemoglobin* production and increased total hemoglobin concentration as much as 5 g/dl over baseline eliminating transfusion requirement in two patients. Parallel declines in circulating nucleated red cell count, and concentrations of serum transform receptor and erythropoietin, are consistent with more effective erythropoiesis. Over extended periods of treatment, no *induction* of other *fetal* proteins and no adverse effects were observed. Particular disease mutations and other genetic factors may be of prime importance in determining the response to agents that *induce* production of *fetal* *hemoglobin*.

13/3,AB/21
DIALOG(R)File 155:MEDLINE(R)

09626638 98055566 PMID: 9395188
BFU-E colony growth in response to hydroxyurea: correlation between in vitro and in vivo *fetal* *hemoglobin* *induction*. Yang Y M; Pace B; Kitchens D; Ballas S K; Shah A; Baliga B S Department of Pediatrics and Comprehensive Sickle Cell Center, University of South Alabama College of Medicine, Mobile 36604, USA.

American journal of hematology (UNITED STATES) Dec 1997, 56 (4) p252-8, ISSN 0361-8609 Journal Code: 7610369
Contract/Grant No.: HL 38632; HL; NHLBI; HL 38639; HL; NHLBI Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Patients with sickle-cell anemia treated with hydroxyurea may have significant reduction in frequency and severity of pain episodes. However, previous clinical trials show a variable response to hydroxyurea. Criteria which can be used to select patients who are likely to respond to hydroxyurea treatment would be useful. Our laboratory has previously demonstrated an inverse linear relationship between the total number of burst-forming unit-erythroid (BFU-E) colonies and *fetal* *hemoglobin* levels in sickle-cell patients treated with hydroxyurea. In the present report, an in vitro cell culture system was established to evaluate the effects of hydroxyurea on BFU-E colony growth and *induction* of *fetal* *hemoglobin* production. Five Hb SS patients who were not previously treated with hydroxyurea and three Hb SS patients who failed to respond to hydroxyurea treatment were included in the study. The results show that the number of BFU-E colonies is decreased from 153.7 to 7.2 per 3 x 10(5) mononuclear cells, whereas *fetal* *hemoglobin* levels were increased from 5.1 to 19.4% in the presence of hydroxyurea in vitro in cultured erythroid progenitors, which were derived from 5 patients before treatment. The number of BFU-E colonies decreased from 153.7 to 2.0 per 3 x 10(5) mononuclear cells in the in vitro cultures obtained from serial peripheral blood samples over a 9- to 20-week period of oral hydroxyurea therapy. A simultaneous rise in *fetal* *hemoglobin* level from 10.2 to 28.6% in the peripheral blood over the same period of hydroxyurea therapy was also observed. Our results demonstrate that the increase in *fetal* *hemoglobin* levels in cells treated with hydroxyurea in vitro is comparable to the rise of *fetal* *hemoglobin* production following hydroxyurea therapy in these patients. On the contrary, these findings were not observed in three previously non-responsive sickle-cell patients. These results suggest that the changes in number of BFU-E colonies and *fetal* *hemoglobin* levels after in vitro exposure to hydroxyurea may be a useful approach to select sickle-cell patients who will respond to hydroxyurea therapy.

13/3,AB/22
DIALOG(R)File 155:MEDLINE(R)

09611602 98039249 PMID: 9371980
Butyrate in the treatment of sickle cell disease and beta-thalassemia. Faller D V; Perrine S P
Boston University School of Medicine, Massachusetts, USA. Current opinion in hematology (UNITED STATES) Mar 1995, 2 (2) p109-17, ISSN 1065-6251 Journal Code: 9430802
Document type: Journal Article; Review; Review, Tutorial
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
The search for, and discovery of, a physiologic model in which the developmentally regulated switch from fetal to adult globin gene expression could be prevented has resulted in the development of a new class of therapeutic agents, consisting of simple fatty acids, such as butyric acid, for the treatment of the beta-hemoglobinopathies. Butyrate and related drugs stimulate fetal (gamma-) globin gene expression in erythroid cells cultured from patients, and in chicken, ovine, and primate animal models. The butyrates are perhaps the first class of drugs designed to transcriptionally activate specific genes--in this particular case, to reactivate the developmentally silenced fetal globin genes. Phase I-II clinical trials resulting from this basic research have been initiated on a small scale during the past 3 years. Analysis of two butyrate-derived therapeutic agents, one delivered intravenously and one orally, has shown initial efficacy in stimulating *fetal* *hemoglobin* expression in 50% to 85% of patients. Correction of the anemia from the beta-hemoglobinopathy has followed *induction* of *fetal* globin, and has been adequate to eliminate the need for erythrocyte transfusions in some patients with beta-thalassemia. These compounds have been relatively safe and without generalized cytotoxicity in patients, but drug tolerance develops in some patients after prolonged therapy. Third-generation, small two- to five-carbon butyrate derivatives are in development. The molecular basis for butyrate action is being defined. Binding of putative regulatory proteins to a specific region of the gamma-globin promoter is altered in vivo in patients receiving butyrate therapy. Further analysis of the mode of action may contribute to development of other therapeutic agents designed to regulate gene transcription.

13/3,AB/23
DIALOG(R)File 155:MEDLINE(R)

09533764 97436573 PMID: 9292546
Induction of *gamma*-globin by histone deacetylase inhibitors. McCaffrey P G; Newsome D A; Fibach E; Yoshida M; Su M S
Molecular Biology Department, Vertex Pharmaceuticals Inc, Cambridge, MA 02139, USA.
Blood (UNITED STATES) Sep 1 1997, 90 (5) p2075-83, ISSN 0006-4971 Journal Code: 7603509

Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

The short-chain fatty acid butyrate has been shown to elevate *fetal* *hemoglobin* (HbF) by *inducing* expression of the *gamma* -globin gene. Regulation of gene expression by butyrate is thought to proceed via inhibition of the enzyme histone deacetylase, leading to elevated levels of core histone acetylation which affect chromatin structure and transcription rates. To determine whether changes in histone acetylation are critical for the regulation of the gamma-globin gene, we tested three potent and specific inhibitors of histone deacetylase, the cyclic tetrapeptides trapoxin and Helminthsporium carbonum toxin (HC toxin), and the antifungal antibiotic trichostatin A for their ability to *induce* *fetal* *hemoglobin* expression in erythroid cells. These compounds *induced* *fetal* *hemoglobin* in both primary erythroid cell cultures and human erythroleukemia (K562) cells. A butyrate-responsive element spanning the duplicated CCAAT box region of the gamma-globin promoter has been identified in transient transfection assays using a reporter construct in K562 cells, and we show that the same promoter region is required for response to trapoxin and trichostatin. Mutational analysis of the gamma-globin promoter indicates that the distal CCAAT box and 3' flanking sequence (CCAATAGCC) is critical for activation by butyrate, trapoxin, and trichostatin, whereas the proximal element (CCAATAGTC) plays a less important role. These results show that inhibition of histone deacetylase can lead to transcriptional activation of gamma-globin promoter reporter gene constructs through proximal promoter elements, and suggest that butyrate *induces* *gamma* -globin expression via such changes in histone acetylation.

13/3,AB/24
DIALOG(R)File 155:MEDLINE(R)

09182264 97085870 PMID: 8931955
Erythroid progenitor proliferation is stimulated by phenoxyacetic and phenylalkyl acids.
Torkelson S; White B; Faller D V; Phipps K; Pantazis C; Perrine S P Hemoglobinopathy-Thalassernia Research Unit, Boston University School of Medicine, MA 02118, USA.
Blood cells, molecules & diseases (UNITED STATES) 1996, 22 (2) p150-8, ISSN 1079-9796 Journal Code: 9509932
Contract/Grant No.: HL-15157; HL; NHLBI; HL-37118; HL; NHLBI Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

Short-chain fatty acids, such as butyrate and propionate, are under investigation as therapeutic stimulants of *fetal* *hemoglobin* production in the beta-hemoglobin disorders. Significant limitations to these fatty acids and derivatives as optimal therapeutics are their rapid metabolism in vivo and their *induction* of cell growth arrest in the G1 phase of the cell cycle. This antiproliferative activity is related to their inhibition of metabolic transport pumps which are essential for cell proliferation. Other small carbon compounds, the phenylalkyl acids, phenoxyacetic acids, and phenylacetic acids, which are structurally resistant to oxidative metabolism, are shown here to *induce* *fetal* globin production in human erythroid cultures at concentrations of 0.2 mM, lower than those required for most other fatty acids. Certain of these compounds were found not to inhibit cellular neutral amino acid transport function in erythroid cells, nor to inhibit erythroid colony (Bfu-e) growth. Certain of these compounds even stimulated human Bfu-e proliferation in vitro beyond that induced by optimal concentrations of hematopoietic growth factors. The combination of increased fetal globin chain production by these compounds and their stimulatory effects on erythropoiesis result in an increase in Hb F-expressing erythroid cells in culture several-fold greater than that achieved by the butyrates. These new compounds thus have the potential to provide superior therapy for the beta-hemoglobinopathies and other anemias.

13/3,AB/25
DIALOG(R)File 155:MEDLINE(R)

08669600 96017259 PMID: 7579419
Stimulation of *fetal* *hemoglobin* production by short chain fatty acids.
Liakopoulou E; Blau C A; Li Q; Josephson B; Wolf J A; Fournarakis B; Raisys V; Dover G; Papayannopoulou T; Stamatoyannopoulos G Department of Medicine, University of Washington, Seattle. Blood (UNITED STATES) Oct 15 1995, 86 (8) p3227-35, ISSN 0006-4971 Journal Code: 7603509
Contract/Grant No.: HL20899; HL; NHLBI; RR00166; RR; NCRR Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

Butyrate, a four-carbon fatty acid, and its two-carbon metabolic product, acetate, are *inducers* of *gamma* -globin synthesis. To test whether other short-chain fatty acids share this property, we first examined whether propionic acid, a three-carbon fatty acid that is not catabolized to acetate, *induces* *gamma*-globin expression. Sodium propionate increased the

frequency of *fetal* *hemoglobin* containing erythroblasts and the gamma/gamma + beta mRNA ratios in adult erythroid cell cultures and F reticulocyte production in a nonanemic juvenile baboon. Short-chain fatty acids containing five (pentanoic), six (hexanoic), seven (heptanoic), eight (octanoic), and nine (nonanoic) carbons *induced* *gamma* -globin expression (as measured by increase in gamma-positive erythroblasts and gamma/gamma + beta mRNA ratios) in adult erythroid burst-forming unit cultures. There was a clear-cut relationship between the concentration of fatty acids in culture and the degree of *induction* of *gamma*-globin expression. Three-, four-, and five-carbon fatty acids were better *inducers* of *gamma* globin in culture as compared with six- to nine-carbon fatty acids. These results suggest that all short-chain fatty acids share the property of *gamma*-globin gene *inducibility* . The fact that valproic acid, a derivative of pentanoic acid, also *induces* *gamma* -globin expression suggests that short-chain fatty acid derivatives that are already approved for human use may possess the property of *gamma*-globin *inducibility* and may be of therapeutic relevance to the beta-chain hemoglobinopathies.

13/3,AB/26
DIALOG(R)File 155:MEDLINE(R)

08368626 95124337 PMID: 7529873
Effects of butyrate and glucocorticoids on gamma- to beta-globin gene switching in somatic cell hybrids.
Zitnik G; Peterson K; Stamatoyannopoulos G; Papayannopoulou T Department of Medicine, University of Washington, Seattle 98195. Molecular and cellular biology (UNITED STATES) Feb 1995, 15 (2) p790-5, ISSN 0270-7306 Journal Code: 8109087
Contract/Grant No.: DK 30852; DK; NIDDK; HL 20899; HL; NHLBI Erratum in Mol Cell Biol 1995 Jun;15(6) 3461
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Butyrate and its analogs have been shown to *induce* *fetal* *hemoglobin* in humans and primates and in erythroid cell cultures. To obtain insights concerning the cellular mechanisms of butyrate action, we analyzed the effects of butyrate on human globin gene expression in hybrids produced by fusing mouse erythroleukemia cells (MEL) with human fetal erythroid cells (HFE). These hybrids initially express human *fetal* *hemoglobin* but subsequently switch to adult globin expression after several weeks in culture. We found that alpha-aminobutyric acid, a butyrate analog which does not induce terminal maturation, strikingly delays the rate of the gamma- to beta-globin gene (gamma-to-beta) switch in the HFE x MEL hybrids. The effect of butyrate on globin expression is transient, with the result that the delay of globin gene switching requires the continuous presence of this compound in culture. Furthermore, butyrate fails to *induce* *fetal* *hemoglobin* expression in hybrids which have switched, suggesting that the effect of this compound on gamma-globin expression is due to inhibition of gamma gene silencing rather than to *induction* of *gamma* gene transcription. Since in other cellular systems, glucocorticoids antagonize the action of butyrate, the effect of dexamethasone on the gamma-to-beta switch in HFE x MEL hybrids was examined. Dexamethasone strikingly accelerated the gamma-to-beta switch, and its effect was irreversible. The effects of dexamethasone and butyrate on the gamma-to-beta switch of the HFE x MEL hybrids appear to be codominant. These results indicate that steroids can have a direct effect on globin gene switching in erythroid cells.

13/3,AB/27
DIALOG(R)File 155:MEDLINE(R)

08328015 95086232 PMID: 7527673
alpha-Amino butyric acid cannot reactivate the silenced gamma gene of the beta locus YAC transgenic mouse.
Pace B; Li Q; Peterson K; Stamatoyannopoulos G Department of Medicine, University of Washington, Seattle 98195. Blood (UNITED STATES) Dec 15 1994, 84 (12) p4344-53, ISSN 0006-4971 Journal Code: 7603509
Contract/Grant No.: HL 20899; HL; NHLBI
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Butyric acid, a naturally occurring fatty acid, has been shown to increase *fetal* *hemoglobin* in BFUe cultures, in primates, and in patients with beta chain hemoglobinopathies. The precise mechanism of *gamma* gene *induction* by butyrate is unknown. Butyrate may *induce* *fetal* *hemoglobin* production in vivo by reactivation of silenced gamma globin genes, by inhibiting the silencing of gamma genes, or by both mechanisms. We examined the effects of butyrate on gamma gene expression in transgenic mice carrying three types of constructs: microLCRA gamma mice, which continue to express the gamma gene in the adult stage of development at a level of one-third to one-fifth of the expression in the fetus; microLCRA gamma psi beta delta beta mice, which display correct developmental regulation of gamma and beta human globin genes and have low level gamma globin expression in the adult; and beta locus YAC mice, which display correct developmental regulation of epsilon, gamma, and

beta globin genes and have a totally silenced gamma gene in the adult stage. Animals were treated with a continuous infusion of alpha-amino butyric acid (alpha-ABA) for 7 days. In microLCRA gamma mice alpha-ABA produced up to a 43-fold *induction* of *gamma* and 9-fold *induction* of mouse alpha globin genes. In contrast, butyrate did not *induce* *gamma* globin expression in the beta locus YAC mice. However, the gamma globin genes of beta locus YAC mice were activated after administration of 5-azacytidine (5-azaC), and the level of gamma globin expression was further increased by administration of alpha-ABA. These results suggest that butyrate cannot reactivate a totally silenced *gamma* gene and that *induction* of *fetal* *hemoglobin* by this compound may require the presence of preactivated gamma globin genes.

13/3,AB/28
DIALOG(R)File 155:MEDLINE(R)

08277416 95036362 PMID: 7524768
Fetal *hemoglobin* *induction* by acetate, a product of butyrate catabolism.
Stamatoyannopoulos G; Blau C A; Nakamoto B; Josephson B; Li Q; Liakopoulou E; Pace B; Papayannopoulou T; Brusilow S W; Dover G Department of Medicine, University of Washington, Seattle 98195.
Blood (UNITED STATES) Nov 1 1994, 84 (9) p3198-204, ISSN 0006-4971 Journal Code: 7603509
Contract/Grant No.: DK45365; DK; NIDDK; HL20899; HL; NHLBI; HL28028; HL; NHLBI; +
Comment in Blood. 1995 Jun 15;85(12) 3765-6; Comment in PMID 7540074 Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Butyrate *induces* *fetal* *hemoglobin* (HbF) synthesis in cultures of erythroid progenitors, in primates, and in man. The mechanism by which this compound stimulates gamma-globin synthesis is unknown. In the course of butyrate catabolism, beta oxidation by mitochondrial enzymes results in the formation of two acetate molecules from each molecule of butyrate. Studies were performed to determine whether acetate itself induces HbF synthesis. In erythroid burst-forming unit (BFU-E) cultures from normal persons, and individuals with sickle cell disease and umbilical-cord blood, dose-dependent increases in gamma-globin protein and gamma mRNA were consistently observed in response to increasing acetate concentrations. In BFU-E cultures from normal adults and patients with sickle cell disease, the ratio of gamma/gamma + beta mRNA increased twofold to fivefold in response to acetate, whereas the percentage of BFU-E progeny staining with an anti-gamma monoclonal antibody (MoAb) increased approximately twofold. Acetate-*induced* increases in *gamma*-gene expression were also noted in the progeny of umbilical cord blood BFU-E, although the magnitude of change in response to acetate was less because of a higher baseline of gamma-chain production. The effect of acetate on HbF *induction* in vivo was evaluated using transgenic mouse and primate models. A transgenic mouse bearing a 2.5-kb mu locus control region (mu LCR) cassette linked to a 3.3-kb A gamma gene displayed a near twofold increase in gamma mRNA during a 10-day infusion of sodium acetate at a dose of 1.5 g/kg/d. Sodium acetate administration in baboons, in doses ranging from 1.5 to 6 g/kg/d by continuous intravenous infusion, also resulted in the stimulation of gamma-globin synthesis, with the percentage of HbF-containing reticulocytes (F reticulocytes) approaching 30%. Surprisingly, a dose-response effect of acetate on HbF *induction* was not observed in the baboons, and HbF *induction* was not sustained with prolonged acetate administration. These results suggest that both two-carbon fatty acids (acetate) and four-carbon fatty acids (butyrate) stimulate synthesis of HbF in vivo.

13/3,AB/29
DIALOG(R)File 155:MEDLINE(R)

08277358 95036304 PMID: 7949149
Augmentation of gamma-globin gene promoter activity by carboxylic acids and components of the human beta-globin locus control region. Safaya S; Ibrahim A; Rieder R F
Department of Medicine, State University of New York Health Science Center, Brooklyn.
Blood (UNITED STATES) Dec 1 1994, 84 (11) p3929-35, ISSN 0006-4971 Journal Code: 7603509
Contract/Grant No.: DK-12401; DK; NIDDK
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Butyric acid increases *fetal* *hemoglobin* synthesis in adult animals and in erythroid cells in culture and *induces* the *gamma* -globin gene promoter in transient expression experiments in K562 cells (McDonagh KT, Nienhuis AW, Blood 78:255a, 1992 [abstr, suppl 1]). We compared the effect of butyrate and other short-chain carboxylic acids in transient expression studies with K562 cells using an expression plasmid bearing a luciferase reporter gene driven by the normal human A gamma-globin gene promoter. Butyrate (4 carbons) increased the activity of the human A gamma-globin gene promoter up to 123 times. Marked augmentation of the normal gamma-promoter activity was also noted with 5-carbon valeric acid (up to 394 times)

and 3-carbon propionic acid (up to 129 times). The branched isobutyric acid as well as phenylacetate showed less ability to increase promoter activity. Addition of the tandemly repeated AP-1/NF-E2 (AP) enhancer sequences from hypersensitive site 2 (HS2) of the locus control region (LCR) increased gamma-promoter activity up to 24 times. Addition of a nearby 16-bp conserved motif (CM) in HS2 (Safaya S, Rieder RF, Blood 78:146a, 1992 [abstr, suppl 1]) to the AP-containing plasmid construct further increased gamma-promoter activity. In the presence of butyrate, the plasmid bearing both the AP and CM sequences showed gene expression up to 477 times greater than that of the basal gamma-promoter-driven luciferase plasmid in the absence of inducer. A plasmid bearing the herpes simplex thymidine kinase promoter was also tested and gene expression was markedly increased by the same organic acids. MEL cells responded to butyrate, valerate, and propionate with *induction* of hemoglobin synthesis. Responses to isobutyrate and 6-carbon caproate required higher concentrations of the compounds. Thus, other short-chain organic acids as well as butyrate increase gamma-promoter activity in the transient expression system, and this activity can be further augmented by incorporating LCR elements into the expression vector. Nonglobin promoters also respond to the same carboxylic acids.

13/3,AB/30
DIALOG(R)File 155:MEDLINE(R)

08201106 94335943 PMID: 7520129
Induction of *fetal* *hemoglobin* in the presence of increased 3-hydroxybutyric acid associated with beta-ketothiolase deficiency.
Galanello R; Cao A; Olivieri N
New England journal of medicine (UNITED STATES) Sep 15 1994, 331 (11) p746-7, ISSN 0028-4793 Journal Code: 0255562
Document type: Letter
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

13/3,AB/31
DIALOG(R)File 155:MEDLINE(R)

08152886 94289722 PMID: 7517215
Induction of *fetal* *hemoglobin* production in subjects with sickle cell anemia by oral sodium phenylbutyrate.
Dover G J; Brusilow S; Charache S
Department of Pediatrics, Johns Hopkins University Medical School, Baltimore, MD.
Blood (UNITED STATES) Jul 1 1994, 84 (1) p339-43, ISSN 0006-4971 Journal Code: 7603509
Contract/Grant No.: HD 11134; HD; NICHD; HD 26358; HD; NICHD; HL 28028; HL; NHLBI; +
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Intravenous arginine butyrate has been shown to increase *fetal* *hemoglobin* (HbF) in sickle cell and thalassemia patients. Recently, we observed that sodium 4-phenylbutyrate, a drug administered orally to treat urea cycle disorders, increases HbF production in nonanemic children and adults. We treated six subjects with sickle cell disease over a period of 14 to 179 days. All subjects received their initial therapy of 9 to 13 g/m2/day as 0.5-g tablets of sodium 4-phenylbutyrate as inpatients. All subjects showed a rapid increase in the percentage of F-reticulocytes (pretreatment, 1% to 20%; posttreatment, 10% to 44%). Four subjects were treated only 11 to 25 days as inpatients. Two of these four subjects failed to respond to the outpatient component because of their inability to maintain an intake of 30 to 40 tablets per day. One subject (C) developed a rash at day 10 and discontinued treatment at day 14. Another subject (B) was transfused for a painful crisis on day 25. Subject A, treated for 179 days, has an increased percentage of F cells, from 54% to 77%, and increased HbF levels, from 10.6% to 18%. Subject F, treated for 154 days, has an increased percentage of F cells, from 59% to 73%, and an increased percentage of HbF, from 10.4% to 16%. All subjects showed some increase in weight. Subject A developed mild transient ankle edema. Myelotoxicity was not seen in any treated patient. Oral administration of sodium 4-phenylbutyrate rapidly increases F-cell production in sickle cell disease.

13/3,AB/32
DIALOG(R)File 155:MEDLINE(R)

08058411 94189535 PMID: 7511330
Transgenic mouse model of pharmacologic *induction* of *fetal* *hemoglobin* : studies using a new ribonucleotide reductase inhibitor, Didox.
Pace B S; Elford H L; Stamatoyannopoulos G
Department of Medicine, University of Washington, Seattle. American journal of hematology (UNITED STATES) Feb 1994, 45 (2) p136-41, ISSN 0361-8609 Journal Code: 7610369
Contract/Grant No.: HL20899; HL; NHLBI
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

Evaluation of pharmacologic agents that stimulate *fetal* *hemoglobin* production has been done mainly in baboons and macaques. We investigated whether results in transgenic mice can predict the stimulation of *fetal* *hemoglobin* in primates, by testing *gamma* globin *induction* in response to a new ribonucleotide reductase inhibitor, Didox. A transgenic mouse line carrying the human A gamma gene linked to a locus control region cassette was used. Treatment of transgenic mice with Didox resulted in *induction* of *gamma* gene expression as documented by an increase in F reticulocytes and F cells and an elevation of gamma/gamma + beta biosynthetic ratio. Similarly, administration of Didox to a baboon in the nonanemic and chronically anemic state resulted in *induction* of *gamma* gene expression as shown by increases in F reticulocytes, F cells, and Hb F. These results suggest that the muLCR-A gamma transgenic mice can be used to screen new pharmacologic compounds for *gamma* globin *inducibility*.

13/3,AB/33
DIALOG(R)File 155:MEDLINE(R)

07932124 94067406 PMID: 7504214
Induction of *fetal* *hemoglobin* with erythropoietin. Blau C A
Nephron (SWITZERLAND) 1993, 65 (2) p336, ISSN 0028-2766 Journal Code: 0331777
Comment on Nephron. 1992;60(3) 371; Comment on PMID 1373475; Comment in Nephron. 1993;65(2):313; Comment in PMID 7504213
Document type: Comment; Letter
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

13/3,AB/34
DIALOG(R)File 155:MEDLINE(R)

07932111 94067393 PMID: 7504213
Induction of *fetal* *hemoglobin* by recombinant human erythropoietin in patients with end-stage renal disease.
Salvati F
Nephron (SWITZERLAND) 1993, 65 (2) p313, ISSN 0028-2766 Journal Code: 0331777
Comment on Nephron. 1993;65(2) 336; Comment on PMID 7504214 Document type: Comment; Letter
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

13/3,AB/35
DIALOG(R)File 155:MEDLINE(R)

07615763 93136445 PMID: 8422469
Fetal *hemoglobin* *induction* with butyric acid: efficacy and toxicity.
Blau C A; Constantoulakis P; Shaw C M; Stamatoyannopoulos G Division of Medical Oncology, University of Washington, Seattle. Blood (UNITED STATES) Jan 15 1993, 81 (2) p529-37, ISSN 0006-4971 Journal Code: 7603509
Contract/Grant No.: CA 09515; CA; NCI; HL 20899; HL; NHLBI Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Butyric acid *induces* *fetal* *hemoglobin* (HbF), a property of potential therapeutic advantage in patients with disorders of globin chain synthesis. We performed dose escalation studies of this compound in baboons to assess whether clinically significant increases in HbF are achievable, and to define the associated toxicities. Additionally, the effect of butyrate in combination with erythropoietin on HbF *induction* was assessed. HbF *induction* in response to butyrate was dependent on the dose and duration of treatment. Doses of butyrate less than 4 g/kg/d were associated with minimal toxicity (hypokalemia) and significant HbF *induction* in these nonanemic animals, with 1 g/kg/d producing an increase in HbF-containing reticulocytes (F reticulocytes) from 0.9% to 8.7% and an increase in HbF from 0.8% to 1.4%. A dose of 2 g/kg/d resulted in an increase in F reticulocytes from 2.1% to 27.8% and an increase in HbF from 0.7% to 2.2%. Doses of 4 g/kg/d in another animal produced an increase in F reticulocytes from 1% to 21.6% and in HbF from 1.9% to 5.3%. Infusions in excess of 4 g/kg/d were complicated (after a variable amount of time) by a decreased level of alertness (caused by hyperosmolality or butyrate itself) and hematologic toxicity (with declines in reticulocyte, white blood cell, and platelet counts). Prolonged infusions of high doses of butyrate (8 to 10 g/kg/d) were associated with peak F reticulocyte percentages reaching 38% to 64.5% and HbF reaching levels in excess of 20%. These high doses (8 to 10 g/kg/d) were complicated in two animals with a striking and unique neuropathologic picture and, in one animal, multiorgan system failure. Erythropoietin in combination with butyrate, induced F reticulocytosis in an additive manner. We conclude that butyric acid is a strong inducer of HbF, particularly when administered in combination with erythropoietin. As chronic toxicities remain undefined, patients in future clinical trials of this and similar compounds should be monitored closely for evidence of neurologic toxicity.

13/3,AB/36
DIALOG(R)File 155:MEDLINE(R)

07589161 93112959 PMID: 7678067
Fetal *hemoglobin* in acute and chronic states of erythroid expansion.
Blau C A; Constantoulakis P; al-Khatti A; Spadaccino E; Goldwasser E; Papayannopoulou T; Stamatoyannopoulos G
Division of Medical Oncology, University of Washington, Seattle. Blood (UNITED STATES) Jan 1 1993, 81 (1) p227-33, ISSN 0006-4971 Journal Code: 7603509
Contract/Grant No.: CA09515; CA; NCI; HL20899; HL; NHLBI Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Physiologic principles underlying the differences in *fetal* *hemoglobin* (HbF) *induction* between acute and chronic states of erythroid expansion are poorly understood. Whereas abrupt erythroid expansion is characterized by a high proportion of reticulocytes coexpressing adult and fetal globin (F reticulocytes), HbF levels wane with chronic erythropoietic stimulation. To investigate this phenomenon, we used various schedules of erythropoietin (epo) administration in primates. Acute intravenous epo administration promoted a 2- to 10-fold preferential *induction* of F reticulocytes compared with total reticulocytes. Total reticulocyte and F reticulocyte production were significantly correlated (correlation coefficient .41 to .74). With chronic epo administration, preferential F reticulocyte production was lost, and there was no correlation between reticulocyte and F reticulocyte production (correlation coefficient -.03). The mean percentage of F reticulocytes did not change between acute and chronic schedules of epo administration. The subcutaneous route of high-dose (3,000 U/kg) epo administration was as effective as intravenous administration in the *induction* of HbF. Reticulocyte and F reticulocyte responses to increasing epo doses were found to be saturable. These results suggest that the kinetics rather than absolute levels of reticulocyte and F reticulocyte response form the basis for preferential F reticulocyte *induction* with acute erythropoietic stimulation, and they support the hypothesis that F reticulocytes arise from a relatively rapid pathway of erythroid maturation.

13/3,AB/37
DIALOG(R)File 155:MEDLINE(R)

07458705 92393105 PMID: 1381630
Induction of erythroid differentiation and *fetal* *hemoglobin* production in human leukemic cells treated with phenylacetate.
Samid D; Yeh A; Prasanna P
Clinical Pharmacology Branch, National Cancer Institute, Bethesda, MD 20892.
Blood (UNITED STATES) Sep 15 1992, 80 (6) p1576-81, ISSN 0006-4971 Journal Code: 7603509
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
There is considerable interest in identifying nontoxic differentiation inducers for the treatment of various malignant and nonmalignant blood disorders, including inborn beta-chain hemoglobinopathies. Using the human leukemic K562 cell line as a model, we explored the efficacy of phenylacetate, an amino acid derivative with a low toxicity index when administered to humans. Treatment of K562 cultures with pharmacologically attainable concentrations of phenylacetate resulted in erythroid differentiation, evident by the reduced growth rate and increased hemoglobin production. The effect was time- and dose-dependent, further augmented by glutamine starvation (phenylacetate is known to deplete circulating glutamine in vivo), and reversible upon cessation of treatment. Molecular analysis showed that phenylacetate *induced* *gamma* globin gene expression with subsequent accumulation of the fetal form of hemoglobin (HbF). Interestingly, the addition of phenylacetate to antitumor agents of clinical interest, eg, hydroxyurea and 5-azacytidine, caused superinduction of HbF biosynthesis. The results suggest that phenylacetate, used alone or in combination with other drugs, might offer a safe and effective new approach to treatment of some hematopoietic neoplasms and severe hemoglobinopathies.

13/3,AB/38
DIALOG(R)File 155:MEDLINE(R)

07418100 92351124 PMID: 1379374
Hydroxyurea *induction* of *fetal* *hemoglobin* synthesis in sickle-cell disease.
Dover G J; Charache S
Johns Hopkins University School of Medicine, Baltimore, MD. Seminars in oncology (UNITED STATES) Jun 1992, 19 (3 Suppl 9) p61-6, ISSN 0093-7754 Journal Code: 0420432
Document type: Journal Article; Review; Review, Tutorial
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
In the past 8 years, it has become apparent that some cytotoxic drugs that interfere with DNA replication can reprogram erythroid progenitors to switch from adult hemoglobin to *fetal* *hemoglobin* (HbF) production. Hydroxyurea has now been shown to substantially

increase HbF in patients with sickle cell anemia. Since HbF interferes with sickle hemoglobin polymerization, hydroxyurea may become an important therapeutic agent for patients with sickle cell anemia.

13/3,AB/39
DIALOG(R)File 155:MEDLINE(R)

07295210 92242389 PMID: 1810966
K562 cells: a source for embryonic globin chains.
Bhaumik K
Department of Cell and Molecular Biology, Medical College of Georgia, Augusta 30912-2100.
Journal of chromatography (NETHERLANDS) Nov 15 1991, 571 (1-2) p37-46, ISSN 0021-9673 Journal Code: 0427043
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
A combination of DEAE-cellulose chromatography and reversed-phase high-performance liquid chromatography (HPLC) has been used to devise a method for generating large quantities of embryonic as well as fetal globin chains. The identity of these globin chains was further confirmed by their tryptic peptide mapping. This technique could, therefore, provide a reliable source for these polypeptides for both analytical and immunological purposes. Moreover, the study of human hemoglobin switching, particularly embryonic to fetal, has been greatly hampered by the absence of a suitable model. K562 cells, due to their potential for differential *induction* of embryonic and *fetal* *hemoglobin* synthesis, can thus be used for this purpose and the various hemoglobins produced can then be effectively monitored using this method.

13/3,AB/40
DIALOG(R)File 155:MEDLINE(R)

07140979 92087381 PMID: 1721467
A transgenic mouse model for studying the *induction* of *fetal* *hemoglobin* in the adult.
Constantoulakis P; Costantini F; Josephson B; Fry D; Mangahas L; Stamatoyannopoulos G
Department of Medicine, University of Washington, Seattle 98195. Transactions of the Association of American Physicians (UNITED STATES) 1990, 103 p80-9, ISSN 0066-9458 Journal Code: 7506109
Contract/Grant No.: DK-31232; DK; NIDDK; HL-20899; HL; NHLBI Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

13/3,AB/41
DIALOG(R)File 155:MEDLINE(R)

06857508 91159639 PMID: 1705838
Locus control region-A gamma transgenic mice: a new model for studying the *induction* of *fetal* *hemoglobin* in the adult. Constantoulakis P; Josephson B; Mangahas L; Papayannopoulou T; Enver T; Costantini F; Stamatoyannopoulos G
Department of Medicine, University of Washington, Seattle 98195. Blood (UNITED STATES) Mar 15 1991, 77 (6) p1326-33, ISSN 0006-4971 Journal Code: 7603509
Contract/Grant No.: DK-31232; DK; NIDDK; HL-20899; HL; NHLBI Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
All pharmacologic agents that *induce* *fetal* *hemoglobin* (Hb) have been discovered with in vivo studies of humans, macaques, and baboons. We tested whether transgenic mice carrying human fetal (gamma) globin genes provide a model for studying the pharmacologic *induction* of HbF in the adult. In initial studies, phenylhydrazine-induced hemolytic anemia, 5-azacytidine, butyrate, or combinations of these treatments failed to activate the human gamma-globin gene in a transgenic mouse line carrying a 4.4-kb G gamma globin gene construct that is expressed only in the embryonic stage of mouse development. Subsequently, adult mice carrying the human A gamma gene linked to the locus control region (LCR) regulatory sequences and expressing heterocellularly HbF (about 25%, gamma-positive cells) were used. Treatments with erythropoietin, 5-azacytidine, hydroxyurea, or butyrate resulted in *induction* of *gamma* gene expression as documented by measurement of F-reticulocytes, the gamma/gamma + beta biosynthetic ratio and the level of steady state gamma mRNA. Administration of erythropoietin or butyrate to transgenic mice carrying a muLCR-beta (human) globin construct, failed to increase human beta-globin expression. These results suggest that the muLCR-A gamma transgenic mice provide a new model for studying the *induction* of *fetal* Hb in the adult.

13/3,AB/42
DIALOG(R)File 155:MEDLINE(R)

06531386 90234870 PMID: 1691937
Fetal calf serum contains activities that *induce* *fetal* *hemoglobin* in adult erythroid cell cultures.
Constantoulakis P; Nakamoto B; Papayannopoulou T; Stamatoyannopoulos G Department of Medicine,

University of Washington, Seattle 98195. Blood (UNITED STATES) May 1 1990, 75 (9) p1862-9, ISSN 0006-4971 Journal Code: 7603509
Contract/Grant No.: HL 20899; HL; NHLBI
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Cultures of peripheral blood or bone marrow erythroid progenitors display stimulated production of *fetal* *hemoglobin* . We investigated whether this stimulation is due to factors contained in the sera of the culture medium. Comparisons of gamma/gamma + beta biosynthetic ratios in erythroid colonies grown in fetal calf serum (FCS) or in charcoal treated FCS (C-FCS) showed that FCS-grown cells had significantly higher gamma/gamma + beta ratios. This increase in globin chain biosynthesis was reflected by an increase in relative amounts of steady-state gamma-globin mRNA. In contrast to its effect on adult cells, FCS failed to influence gamma-chain synthesis in fetal burst forming units-erythroid (BFU-E) colonies. There was a high correlation of gamma-globin expression in paired cultures done with C-FCS or fetal sheep serum. Dose-response experiments showed that the *induction* of *gamma* -globin expression is dependent on the concentration of FCS. These results indicate that FCS contains an activity that *induces* *gamma*-globin expression in adult erythroid progenitor cell cultures.

13/3,AB/43
DIALOG(R)File 155:MEDLINE(R)

06467541 90178554 PMID: 1689967
Induction of *fetal* *hemoglobin* by cell-cycle-specific drugs and recombinant erythropoietin.
Stamatoyannopoulos G; Veith R; al-Khatti A; Papayannopoulou T Department of Medicine, University of Washington, Seattle 98195. American journal of pediatric hematology/oncology (UNITED STATES) Spring 1990, 12 (1) p21-6, ISSN 0192-8562 Journal Code: 7908071 Contract/Grant No.: HL20877; HL; NHLBI
Document type: Journal Article; Review; Review, Tutorial
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
During the last several years, studies in humans and experimental animals have identified several compounds that *induce* *fetal* *hemoglobin* in the adult. These include: cell-cycle-specific drugs, other cytotoxic drugs, butyric acid analogs, and erythropoietin. Several of these compounds *induce* *fetal* *hemoglobin* indirectly by triggering kinetics of rapid erythroid regeneration. High doses of erythropoietin increase the frequency of erythroid progenitors programmed to hemoglobin F. This results in transient increases of hemoglobin F-containing cells (F cells) in the peripheral blood. Erythropoietin and hydroxyurea increase F cells in a cooperative fashion. Although high doses of erythropoietin can induce F cell production in humans, the practical relevance of such observations is unclear

13/3,AB/44
DIALOG(R)File 155:MEDLINE(R)

06446961 90139053 PMID: 2482498
Induction of *fetal* *hemoglobin* in sickle cell patients by hydroxyurea: the N.I.H. experience.
Rodgers G P; Dover G J; Noguchi C T; Schechter A N; Nienhuis A W Laboratory of Chemical Biology, NIDDK, Bethesda, Maryland 20892. Progress in clinical and biological research (UNITED STATES) 1989, 316B p281-93, ISSN 0361-7742 Journal Code: 7605701
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

13/3,AB/45
DIALOG(R)File 155:MEDLINE(R)

06335887 90028718 PMID: 2478217
On the *induction* of *fetal* *hemoglobin* by butyrates: in vivo and in vitro studies with sodium butyrate and comparison of combination treatments with 5-AzaC and AraC.
Constantoulakis P; Knitter G; Stamatoyannopoulos G Department of Medicine, University of Washington, Seattle 98195. Blood (UNITED STATES) Nov 1 1989, 74 (6) p1963-71, ISSN 0006-4971 Journal Code: 7603509
Contract/Grant No.: HL 20899; HL; NHLBI; RR 00166; RR; NCRR Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
To obtain information on the cellular mechanism of *induction* of *fetal* *hemoglobin* (HbF) by sodium butyrate (NaB), we treated adult baboons with NaB and assessed its effects on HbF expression. Infusion of NaB increased F reticulocytes and F-positive CFUe and e-cluster colonies without *induction* of reticulocytosis or increase in progenitor cell numbers. Addition of NaB in bone marrow cultures increased the frequency of F-positive CFUe and e-clusters without increasing progenitor cell numbers. NaB induced HbF in human adult BFUe cultures and increased the gamma/gamma + beta

globin chain and mRNA ratios in short-term incubations of culture-derived erythroblasts. There was a synergistic *induction* of HbF by NaB and 5-azacytidine (5-azaC), but not when the animal was treated with NaB and cytarabine (AraC). Our results suggest that the activation of gamma-globin expression by NaB reflects an action of this compound on globin genes or globin chromatin.

13/3,AB/46
DIALOG(R)File 155:MEDLINE(R)

05689951 88125112 PMID: 2448812
On the *induction* of *fetal* *hemoglobin* in the adult; stress erythropoiesis, cell cycle-specific drugs, and recombinant erythropoietin.
Stamatoyannopoulos G; Veith R; Al-Khatti A; Fritsch E F; Goldwasser E; Papayannopoulou T
Department of Medicine, University of Washington, Seattle. Progress in clinical and biological research (UNITED STATES) 1987, 251 p443-53, ISSN 0361-7742 Journal Code: 7605701
Contract/Grant No.: HL 20899; HL; NHLBI; HL 21676; HL; NHLBI Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

13/3,AB/47
DIALOG(R)File 155:MEDLINE(R)

05517540 87273205 PMID: 2440559
Fetal *hemoglobin* gene activation in a phase II study of 5,6-dihydro-5-azacytidine for bronchogenic carcinoma.
Carr B I; Rahbar S; Doroshow J H; Blayney D; Goldberg D; Leong L; Asmeron Y
Cancer research (UNITED STATES) Aug 1 1987, 47 (15) p4199-201, ISSN 0008-5472 Journal Code: 2984705R
Contract/Grant No.: CA 33572-06; CA; NCI
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
5-Azacytidine and several of its analogues are known to inhibit DNA methylation, alter gene expression, and inhibit cell growth. We report a Phase II study in which we investigated the antineoplastic activity of 5,6-dihydro-5-azacytidine and its *induction* of *fetal* *hemoglobin* synthesis when given by a 5-day continuous i.v. infusion of 1650 mg/m2/day that was repeated every 21 days. *Fetal* *hemoglobin* was measured in all patients; increased synthesis was found in 13 of the 17, in the absence of clinically significant anemia. Of the four patients who did not develop increased *fetal* *hemoglobin* , three had only one cycle of therapy. Fourteen patients with bronchogenic carcinoma were treated, and ten were evaluable for disease response. Five patients had disease stability of 2 or more mo, and five progressed on treatment. Three additional patients with mesothelioma were treated, and the two who were evaluable for disease response had stabilization of their disease. Fifteen of the 17 patients who received 5,6-dihydro-5-azacytidine developed a pleuritic-type chest pain, 12 had abnormal electrocardiograms, and four developed positive anti-nuclear antibodies. No significant hemopoietic, hepatic, or renal toxicities were observed. This study demonstrates that 5,6-dihydro-5-azacytidine in the dose and schedule used has no significant therapeutic activity in the treatment of lung cancer but does possess an unusual spectrum of clinical toxicities as well as the property of *inducing* *fetal* *hemoglobin* synthesis.

13/3,AB/48
DIALOG(R)File 155:MEDLINE(R)

04484893 84172235 PMID: 6200940
Arabinosylcytosine *induces* *fetal* *hemoglobin* in baboons by perturbing erythroid cell differentiation kinetics.
Papayannopoulou T; Torrealba de Ron A; Veith R; Knitter G; Stamatoyannopoulos G
Science (UNITED STATES) May 11 1984, 224 (4649) p617-9, ISSN 0036-8075 Journal Code: 0404511
Contract/Grant No.: GM 15253; GM; NIGMS; HL-07093; HL; NHLBI; HL-20899; HL; NHLBI; +
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Arabinosylcytosine, a compound that inhibits DNA synthesis in rapidly dividing cells, stimulates *fetal* *hemoglobin* in adult baboons and produces significant perturbations in the pools of erythroid progenitors. It appears that changes in the kinetics of erythroid cell differentiation rather than direct action on the gamma genes underlie stimulation of *fetal* *hemoglobin* in the adult animals in vivo. These results also suggest that chemotherapeutic agents selected for their low carcinogenic or mutagenic potential could be used for therapeutic *induction* of *fetal* *hemoglobin* in patients with sickle cell anemia.

13/3,AB/49

DIALOG(R)File 155:MEDLINE(R)

04285837 83295975 PMID: 6193375
Preferential *induction* of *fetal* versus embryonic globin chains in human leukemic cell lines.
Gianni A M; Presta M; Polli E; Peschle C; Lettieri F; Saglio G; Comi P; Giglioni B; Ottolenghi S
Leukemia research (ENGLAND) 1982, 6 (2) p155-63, ISSN 0145-2126 Journal Code: 7706787
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
By use of a newly developed technique combining affinity chromatography of hemoglobin on haptoglobin-Sepharose and IEF of globin chains, we analyzed the globin synthetic pattern of human K562 cells in both the basal state and after addition of several potential inducers. Hemin only was found effective: its addition at 50 microM results in a quantitative increase of globin chain synthesis (from 0.3 to 1% up to 5%) and a qualitative "switch" with a striking increase of alpha and a decrease of epsilon and zeta chains (relative to the prevailing gamma chains). This system, in which hemin induces changes that mimic to some extent the normal embryonic-fetal switch, might therefore provide a cellular model for investigating molecular mechanisms of globin gene regulation. In addition similar results were obtained with a different human myeloid leukemia cell line, the KG1, thus raising the possibility that the expression of embryonic globin genes in malignant cells might not be simply the consequence of abnormal gene expression but rather reflect a possibly physiological differentiation phenomenon.

13/3,AB/50
DIALOG(R)File 155:MEDLINE(R)

03882243 82157192 PMID: 6175208
Identification and quantitation of embryonic and three types of *fetal* *hemoglobin* produced on *induction* of the human pluripotent leukemia cell line K-562 with hemin.
Fuhr J E; Bamberger E; Lozzio C B; Lozzio B B; Felice A E; Altay G; Webber B B; Reese A L; Mayson S M; Huisman T H
American journal of hematology (UNITED STATES) Feb 1982, 12 (1) p1-12, ISSN 0361-8609 Journal Code: 7610369
Contract/Grant No.: CA 18185-06; CA; NCI; HLB 23736; HL; NHLBI; HLB-05168 ; HL; NHLBI; +
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
The hemoglobins synthesized by the pluripotent K-562 leukemia cell line of human origin after *induction* with hemin have been isolated by DEAE-cellulose chromatography and characterized by electrophoresis, high pressure liquid chromatography, and a radioimmunological assay. Six hemoglobin zones have been observed with the following likely compositions. Zone 1: alpha 2 epsilon 2, or HB Gower-2; zone 2: zeta 2 epsilon 2, or HB Gower-1; zone 3: zeta 2 gamma 2, or HB Portland-I; zone 4: Hb F, or alpha 2 gamma 2; zone 5: a mixture of acetylated HB Portland-I and Hb F; zone 6: Hb Bart's, or gamma 4. The embryonic Hbs (zones 1, 2, and 3) constituted 50%-75% of the total Hb present; the quantities varied from one experiment to the other. Both Hb Gower-1 and Hb Gower-2 were present. The gamma chain was heterogeneous and contained the G gamma, A gamma I, and A gamma T types in a ratio of about 4:2:1, indicating a heterozygosity for the Ile leads to Thr substitution at position gamma 75. The methodology used can be applied for additional studies evaluating quantitative changes in Hb types due to in vitro manipulations.
? s photoreceptor()cell()survival
7871 PHOTORECEPTOR
1605728 CELL
314780 SURVIVAL
S14 13 PHOTORECEPTOR()CELL()SURVIVAL
? t s14/3,ab/all

14/3,AB/1
DIALOG(R)File 155:MEDLINE(R)

13972055 22242540 PMID: 12355060
Alterations in retinal rod outer segment fatty acids and light-damage susceptibility in P23H rats.
Bicknell Ina Rea; Darrow Ruth; Barsalou Linda; Fliesler Steven J; Organisciak Daniel T; et al
Petticrew Research Laboratory, Department of Biochemistry and Molecular Biology, School of Medicine, Wright State University, Cox Institute, Dayton, OH 45429, USA. ina.bicknell@wright.edu
Molecular vision electronic resource (United States) Sep 5 2002, 8 p333-40, ISSN 1090-0535 Journal Code: 9605351
Contract/Grant No.: EY01959; EY; NEI; EY07361; EY; NEI; + Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
PURPOSE: To determine whether dietary-induced alterations in the long-chain polyunsaturated fatty acid content of retinal rod outer segments (ROS) of P23H rats, a transgenic model of retinitis pigmentosa (RP), prolongs photoreceptor cell life. METHODS: Heterozygous P23H and normal Sprague-Dawley rats were fed a standard house diet or a diet deficient in

18:3n-3. Diet-deficient rats were given supplements of either linseed oil (high in 18:3n-3) or fish oil (high in 20:5n-3). ROS fatty acid profiles and serum fatty acids were determined by gas chromatography. Serum cholesterol was evaluated by HPLC. Retinal damage was assessed by measuring whole-retina rhodopsin and DNA content before and after exposure to high-intensity light. RESULTS: The retinas of 60 day old, cyclic-light-reared, P23H transgenic rats contained 50% of the rhodopsin and 75% of the DNA content found in control Sprague-Dawley rats. Eight hours of intense light had little effect on the rhodopsin or DNA content in the Sprague-Dawley rats, but resulted in rhodopsin and DNA losses of nearly 70%, compared to controls, in P23H animals fed either a standard or an 18:3n-3-deficient diet. Supplementation with linseed oil resulted in small, statistically insignificant, increases in the rhodopsin and DNA losses, which occurred after exposure to intense light, in P23H transgenics. In unexposed animals, supplementation with linseed oil or fish oil had no effect on either rhodopsin or DNA levels in P23H rats or in Sprague-Dawley controls. On standard diet, the ROS 22:6n-3 (DHA) content in P23H rats was lower than that of control animals. DHA decreased in both groups when an 18:3-deficient diet was fed. The reduction was greater in controls than in P23H transgenics, but a concomitant increase in 22:5n-6 was nearly the same in both groups. Supplementation of the 18:3-deficient diet with linseed oil or fish oil in P23H rats resulted in a ROS fatty acid profile comparable to that of Sprague-Dawley rats raised on a standard diet. Serum DHA and 22:5n-6 levels were low in both groups. No significant differences in serum cholesterol were observed as a function of genotype or diet. CONCLUSIONS: Heterozygous P23H rats are capable of forming ROS DHA from dietary fatty acid precursors found in linseed oil (18:3n-3) or fish oil (20:5n-3). Under all dietary conditions, P23H transgenics are highly susceptible to retinal damage from exposure to intense light. Although levels of DHA in the ROS of P23H rats could be altered by dietary manipulation, only small changes in *photoreceptor* *cell* *survival*, as measured by whole-retina rhodopsin and DNA content, were observed. The lower-than-normal levels of ROS DHA may reflect an adaptive, possibly protective, mechanism in the P23H transgenic rat model of RP.

14/3,AB/2
DIALOG(R)File 155:MEDLINE(R)

13917152 22119649 PMID: 12123634
Subretinal transplantation of brain-derived precursor cells to young RCS rats promotes *photoreceptor* *cell* *survival*. Wojciechowski Anita Blixt; Englund Ulrica; Lundberg Cecilia; Wictorin Klas; Warfvinge Karin; et al
Department of Ophthalmology, Wallenberg Retina Center, Lund University Hospital, S-221 84, Lund, Sweden.
Experimental eye research (England) Jul 2002, 75 (1) p23-37, ISSN 0014-4835 Journal Code: 0370707
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
The potential use of in vitro-expanded precursor cells or cell lines in brain repair includes transplantation of such cells for cell replacement purposes and the activation of host cells to provide 'self-repair'. Recently, it has been reported that the immortalized brain-derived cell line RN33B (derived from the embryonic rat medullary raphe) survive, integrate and differentiate after subretinal grafting to normal adult rats. Here, it is demonstrated that grafts of these cells survive for at least 6 weeks after implantation into postnatal days 21 and 35 retinas of normal and Royal College of Surgeons rats, a model of retinal degeneration. Implanted cells integrate into the retinal pigment epithelium and the inner retinal layers, and the anterior part of the optic nerve of both normal and Royal College of Surgeons rats. The RN33B cells migrate within the retina, occupying the whole retina from one eccentricity to the other. A significant number of the grafted cells differentiate into glial cells, as shown by the double labelling of the reporter genes LacZ or green fluorescent protein, with several glial markers, including oligodendrocytic markers. Many implanted cells in the host retina were in a proliferative stage judging from proliferative cell nuclear antigen and SV40 large T-antigen immunohistochemistry. Interestingly, there was a promotion of photoreceptor survival, extending over more than 2/3 of the superior hemisphere, in Royal College of Surgeons rats transplanted at postnatal day 21, but not at postnatal day 35. In addition, grafted cells were found in the surviving photoreceptor layer in these rats.

14/3,AB/3
DIALOG(R)File 155:MEDLINE(R)

13101133 21844088 PMID: 11853757
The carboxyl-terminal domain is essential for rhodopsin transport in rod photoreceptors.
Concepcion Francis; Mendez Ana; Chen Jeannie
The Mary D. Allen Laboratory for Vision Research, Beckman Macular Research Center, Doheny Eye Institute and Department of Ophthalmology, Keck School of Medicine, University of Southern California, 1333 San Pablo Street, Los Angeles, CA 90089-9112, USA.
Vision research (England) Feb 2002, 42 (4) p417-26, ISSN 0042-6989 Journal Code: 0417402
Contract/Grant No.: EY03040; EY; NEI; EY12155; EY;

NEI
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
The role of the carboxyl-terminal domain in rhodopsin transport was investigated using transgenic mice expressing a rhodopsin truncation mutant lacking the terminal 15 amino acids (S334ter). It was previously shown that S334ter translocates to the outer segment in the presence of endogenous rhodopsin. We now show that in the absence of endogenous rhodopsin S334ter mis-localizes to the plasma membrane and fails to reconstitute outer segment structures. Surprisingly, this mis-localization does not affect *photoreceptor* *cell* *survival* . These results provide further evidence on the important role of the COOH-terminal domain in rhodopsin trafficking and demonstrate an absolute requirement of this domain for correct vectorial transport of rhodopsin in rod photoreceptors.

14/3,AB/4
DIALOG(R)File 155:MEDLINE(R)

11351035 21419308 PMID: 11527926
Isolation and characterization of galectins in the mammalian retina. Uehara F; Ohba N; Ozawa M
Department of Ophthalmology, Kagoshima University Faculty of Medicine, Japan. fuehara@med5.kufm.kagoshima-u.ac.jp
Investigative ophthalmology & visual science (United States) Sep 2001, 42 (10) p2164-72, ISSN 0146-0404 Journal Code: 7703701 Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
PURPOSE: Previous studies have suggested that galectins may be involved in retinal adhesion and *photoreceptor* *cell* *survival*. To elucidate the underlying mechanisms, the authors isolated retinal galectins, determined their types and distributions, and investigated the validity of the hypothesis, using rat models. METHODS: An antibody was prepared against a bovine retinal lectin that was isolated by use of a lactose-agarose column. cDNA of the lectin was isolated by screening of a bovine retinal cDNA library, using the antibody, and then was sequenced. The cDNAs of rat retinal galectins were also isolated by means of polymerase chain reaction and used to produce an antibody against recombinant galectin-3. Using the described antibodies, the authors examined the distributions of galectins in bovine and rat retinas, morphologic changes of rat retinas induced by the antibodies, and distributional changes of galectins in constant-light-exposed rat retinas. RESULTS: The cDNAs of bovine galectin-1, rat galectin-1, and rat galectin-3 were isolated. Galectin-1 was found in various regions, including the retinal pigment epithelium, outer limiting membrane, and outer plexiform layer in bovine and rat retinas. Galectin-3 was increasingly detected in the cytoplasm of Muller cells after constant light exposure after an increase in its transcript. Retinal detachment and vacuolation of the outer plexiform layer were induced in rat eyes by intravitreous injection of the anti-galectin-1 antibody. CONCLUSIONS: Galectin-1 may be involved in adhesion of the photoreceptor and outer plexiform layers by interacting with glycoconjugates with beta-galactoside residues in the interphotoreceptor matrix and synaptic cleft matrix. Galectin-3 may increase in Muller cells of a degenerative rat retina, probably through endogenous anti-apoptosis.

14/3,AB/5
DIALOG(R)File 155:MEDLINE(R)

09844273 98270039 PMID: 9602059
RPE secreted proteins and antibody influence *photoreceptor* *cell* *survival* and maturation.
Sheedlo H J; Nelson T H; Lin N; Rogers T A; Roque R S; Turner J E Department of Anatomy and Cell Biology, University of North Texas Health Science Center at Fort Worth 76107, USA. hsheedlo@hsc.unt.edu Brain research. Developmental brain research (NETHERLANDS) Apr 17 1998, 107 (1) p57-69, ISSN 0165-3806 Journal Code: 8908639 Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Proteins in media conditioned by retinal pigment epithelial cells (RPE-CM) and an antibody against these proteins (RPE-SP) were tested for their respective effects on rat retinal development in vitro and in vivo. Proteins of RPE-CM were separated in denaturing gels and evaluated by Western blot analysis. Retinal explants from postnatal day 2 (P2) rats were cultured in RPE-CM only or CM diluted with the RPE-SP antibody and, after 7 days, the explants were dissociated into single cells that were immunostained for opsin. RPE-CM or antibody was also injected into the vitreous of postnatal day 7 (P7) Long-Evans rats and analyzed 7 and 21 days later. Electrophoretic analysis of RPE-CM predominantly showed 60-70 kDa proteins; when these proteins were probed with RPE-SP antibody by Western blot, immunoreactive proteins were restricted to this narrow molecular weight range. In P2 retinal explant cultures supplemented with RPE-CM, long ganglion cell-like neurites were detected in 3 days. This activity was nullified in explant cultures grown in RPE-CM titrated with antibody, and these explants appeared to

degenerate within 5 days. Over 80% of dissociated retinal cells from explants 7 days after treatment with RPE-CM expressed opsin, compared to only 20% of cells from explants grown in defined medium or serum. Retinas of P14 rats injected intravitreally with RPE-CM at P7 had increased numbers of ectopic photoreceptor cells within the inner nuclear layer when compared to retinas of sham-injected eyes. In contrast, retinas of eyes injected intravitreally with RPE-SP antibody exhibited shorter outer (OS) and inner (IS) segments and thinner outer nuclear (ONL) and outer plexiform (OPL) layers than retinas of sham-injected eyes. In conclusion, proteins in RPE-CM appeared to accelerate and maximize the development of rat photoreceptor cells in vitro, while intravitreal injections of its antibody caused an apparent retardation of outer segment maturation. These results suggest that a protein(s) secreted by RPE plays a key role in normal retinal development, particularly in *photoreceptor* *cell* *survival* and outer segment maturation.

14/3,AB/6
DIALOG(R)File 155:MEDLINE(R)

09558337 97477384 PMID: 9334340
The phosphatidylinositol transfer protein domain of Drosophila retinal degeneration B protein is essential for *photoreceptor* *cell* *survival* and recovery from light stimulation.
Milligan S C; Alb J G; Elagina R B; Bankaitis V A; Hyde D R Department of Biological Sciences, University of Notre Dame, Notre Dame, Indiana 46556, USA.
Journal of cell biology (UNITED STATES) Oct 20 1997, 139 (2) p351-63 , ISSN 0021-9525 Journal Code: 0375356
Contract/Grant No.: EY08058; EY; NEI; GM44530; GM; NIGMS Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
The Drosophila retinal degeneration B (rdgB) gene encodes an integral membrane protein involved in phototransduction and prevention of retinal degeneration. RdgB represents a nonclassical phosphatidylinositol transfer protein (PITP) as all other known PITPs are soluble polypeptides. Our data demonstrate roles for RdgB in proper termination of the phototransduction light response and dark recovery of the photoreceptor cells. Expression of RdgB's PITP domain as a soluble protein (RdgB-PITP) in rdgB2 mutant flies is sufficient to completely restore the wild-type electrophysiological light response and prevent the degeneration. However, introduction of the T59E mutation, which does not affect RdgB-PITP's phosphatidylinositol (PI) and phosphatidycholine (PC) transfer in vitro, into the soluble (RdgB-PITP-T59E) or full-length (RdgB-T59E) proteins eliminated rescue of retinal degeneration in rdgB2 flies, while the light response was partially maintained. Substitution of the rat brain PITPalpha, a classical PI transfer protein, for RdgB's PITP domain (PITPalpha or PITPalpha-RdgB chimeric protein) neither restored the light response nor maintained retinal integrity when expressed in rdgB2 flies. Therefore, the complete repertoire of essential RdgB functions resides in RdgB's PITP domain, but other PITPs possessing PI and/or PC transfer activity in vitro cannot supplant RdgB function in vivo. Expression of either RdgB-T59E or PITPalpha-RdgB in rdgB+ flies produced a dominant retinal degeneration phenotype. Whereas RdgB-T59E functioned in a dominant manner to significantly reduce steady-state levels of rhodopsin, PITPalpha-RdgB was defective in the ability to recover from prolonged light stimulation and caused photoreceptor degeneration through an unknown mechanism. This in vivo analysis of PITP function in a metazoan system provides further insights into the links between PITP dysfunction and an inherited disease in a higher eukaryote.

14/3,AB/7
DIALOG(R)File 155:MEDLINE(R)

09177468 97079536 PMID: 8921247
Photoreceptor repair in response to RPE transplants in RCS rats: outer segment regeneration.
Lin N; Fan W; Sheedlo H J; Aschenbrenner J E; Turner J E Department of Anatomy and Cell Biology, North Texas Eye Research Institute, University of North Texas Health Science Center, Fort Worth 76107, USA.
Current eye research (ENGLAND) Oct 1996, 15 (10) p1069-77, ISSN 0271-3683 Journal Code: 8104312
Contract/Grant No.: EY-04337; EY; NEI
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
PURPOSE: We have previously shown that transplants of normal rat neonatal RPE cells rescued photoreceptor cells in retinas of Royal College of Surgeons (RCS) dystrophic rats for up to one year. In this study, we investigated the photoreceptor rescue effects in RCS rats within the first three weeks following transplantation in an attempt to determine if RPE transplants initiate repair mechanisms, specifically, outer segment (OS) regeneration. METHODS: Freshly isolated RPE cells from neonatal pigmented Long Evans rats were transplanted into the subretinal space of 22-23 day-old RCS rats using a transscleral approach. For controls, vehicle was similarly injected. RESULTS:

When analyzed at 10 days post-transplantation, long inner segments were observed with short buds of outer segment growth in the area of the RPE-cell transplants. The outer segments were of insufficient length to be measured at 10 days, but by 14 and 21 days, OS were 2.02 +/- 0.32 microns and 18.80 +/- 2.78 microns, respectively. In vehicle-injected retinas from 10 to 21 days postsurgery, outer segments were not observed and the inner segments were three-fold shorter than in RPE-transplanted retinas. At 10 days post-transplantation, most RPE cells were seen in the subretinal space, but a few had attached to Bruch's membrane; however, by 21 days, many of the transplanted RPE cells had attached to Bruch's membrane, although a few were found free in the subretinal space. CONCLUSIONS: This study has shown that transplants of normal rat neonatal RPE cells have the capacity to support not only *photoreceptor* *cell* *survival* but also initiate early repair mechanisms as exhibited by outer segment regeneration in RCS retinas. These results also conclusively show the important role that the RPE plays in outer segment growth and maturation.

14/3,AB/8
DIALOG(R)File 155:MEDLINE(R)

09104759 97000866 PMID: 8843922
Survival and regeneration of adult human and other mammalian photoreceptors in culture.
Gaudin C; Forster V; Sahel J; Dreyfus H; Hicks D
Physiopathologie Retinienne INSERM CJF-9202, Strasbourg, France. Investigative ophthalmology & visual science (UNITED STATES) Oct 1996, 37 (11) p2258-68, ISSN 0146-0404 Journal Code: 7703701
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
PURPOSE: Fully mature neurons of central nervous system origin generally are considered unable to survive for extended periods of time in simple culture conditions. The authors report that adult and aged human, porcine, and rodent retinal neurons, including rod and cone photoreceptors, constitute an exception to this idea. METHODS: Cells were dissociated from human postmortem retinas, adult mammalian retinas, and selected brain regions and were seeded into tissue culture plates and left to develop as monolayer cultures for up to 2 months. A battery of antibody markers was used to identify the nature and morphology of the cells in vitro. RESULTS: *Photoreceptor* *cell* *survival* of rods and cones was observed routinely when the delay between the time of death until culture preparation was 50 hours or less, compatible with current eye bank practice. Two-week-old cultures were formed of rod photoreceptors, representing approximately 50% of neuronal cell types; cone photoreceptors, representing 5% to 30% of neuronal cell types; other retinal neurons (especially amacrine cells approximately 20%); and retinal glial cells, present in variable numbers. Glial cells were essential for long-term photoreceptor survival and neurite outgrowth. Adult mammalian brain neurons isolated under the same conditions did not survive. CONCLUSIONS: Fully adult human and other mammalian retinal neurons, including photoreceptors, exhibit remarkable plasticity in vitro, and such monolayer models may have applications in physiological, pharmacologic, and toxicologic studies of human and other mammalian retina.

14/3,AB/9
DIALOG(R)File 155:MEDLINE(R)

08371881 95129805 PMID: 7828824
Mutations in calphotin, the gene encoding a Drosophila photoreceptor cell-specific calcium-binding protein, reveal roles in cellular morphogenesis and survival.
Yang Y; Ballinger D
Graduate Program in Molecular Biology, Cornell University Graduate School of Medical Sciences, New York, New York 10021.
Genetics (UNITED STATES) Oct 1994, 138 (2) p413-21, ISSN 0016-6731 Journal Code: 0374636
Contract/Grant No.: P30-CA-08748-27; CA; NCI
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Calphotin is a Drosophila photoreceptor cell-specific protein expressed very early in eye development, at the time when cell-type decisions are being made. Calphotin is a very hydrophobic and proline-rich protein which lacks obvious transmembrane domains. The cDNA encoding Calphotin was mapped to a region removed by a set of existing chromosomal deletions. Mutations that alter photoreceptor cell structure and development were isolated that fail to complement these deletions. These mutations fall into two classes. Class I mutations alter the structure of the rhabdomere, a photoreceptor cell organelle specialized for phototransduction. Class II mutations have rough eyes, due to misorientation of the rhabdomeres and photoreceptor cell death. Transformation rescue of these phenotypes in transgenic flies bearing calphotin genomic DNA indicates that both classes of mutations are in the calphotin gene. Analysis of these mutations suggest that Calphotin plays important roles in both rhabdomere development and in *photoreceptor* *cell* *survival*.

14/3,AB/10
DIALOG(R)File 155:MEDLINE(R)

08048966 94200318 PMID: 8150027
Effects of RPE age and culture conditions on support of *photoreceptor* *cell* *survival* in transplanted RCS dystrophic rats.
Sheedlo H J; Li L; Turner J E
Department of Anatomy and Cell Biology, University of North Texas Health Science Center, Fort Worth 76107-2690.
Experimental eye research (ENGLAND) Dec 1993, 57 (6) p753-61, ISSN 0014-4835 Journal Code: 0370707
Contract/Grant No.: EY 04337; EY; NEI
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
This study assessed the effects of transplants of freshly isolated or cultured (ie. passaged) retinal pigment epithelial (RPE) cells from neonatal and adult normal and RCS pigmented dystrophic rats on *photoreceptor* *cell* *survival* in retinas of 22-26-day-old pink-eyed RCS dystrophic rats. We determined that retinas of 2-month-old RCS rats transplanted at 26 days with RPE cells of adult RCS rats did not support *photoreceptor* *cell* *survival* above that seen in sham or nontreated control RCS retinas, as outer nuclear layer (ONL) thicknesses were not significantly different (10.0 +/- 1.31 microns, 11.7 +/- 4.04 microns and 9.42 +/- 1.88 microns, respectively). Surprisingly, in this same transplant group, RPE transplants from neonatal RCS dystrophic rats were able to promote *photoreceptor* *cell* *survival* similar to that seen in transplants of neonatal Long Evans rats, as evidenced by similar ONL thicknesses (34.4 +/- 3.16 microns and 33.6 +/- 6.03 microns, respectively), but the rescue effect quickly diminished. However, in retinas of 22-26-day-old RCS rats transplanted with RPE cells from adult Long Evans rats, the level of photoreceptor cell rescue was approximately 48% (ONL: 19.6 +/- 2.79 microns), when compared to retinas transplanted with RPE cells from neonatal Long Evans rats, but significantly greater than that caused by transplants of RPE cells from adult RCS rats.(ABSTRACT TRUNCATED AT 250 WORDS)

14/3,AB/11
DIALOG(R)File 155:MEDLINE(R)

07391814 92324338 PMID: 1385580
RPE conditioned medium stimulates *photoreceptor* *cell* *survival*, neurite outgrowth and differentiation in vitro. Gaur V P; Liu Y; Turner J E
Department of Neurobiology, Bowman Gray School of Medicine, Wake Forest University, Winston-Salem, NC 27013.
Experimental eye research (ENGLAND) May 1992, 54 (5) p645-59, ISSN 0014-4835 Journal Code: 0370707
Contract/Grant No.: EY-04377; EY; NEI
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
In the present study we have investigated retinal pigment epithelium-photoreceptor cell interactions in vitro, and their contributions to *photoreceptor* *cell* *survival* and differentiation. Preparations enriched for intact photoreceptor cells from neonatal rat retina were grown in either serum-free medium supplemented with RPE-conditioned medium (RPE-CM) or in serum-free medium alone. A variety of substrate conditions were tested for the best neurite outgrowth. Cultures were monitored for 7 days by light and electron microscopy, as well as by opsin, vimentin and carbonic anhydrase-C immunocytochemistry. RPE-CM was found to stimulate both proliferation of flat cells and photoreceptor differentiation. The number of photoreceptors bearing neurites and their neurite length measurements showed significant differences between the RPE-CM group and the control group within 20 hr in culture. Elimination of contaminating flat cells by the addition of an antimitotic drug prevented photoreceptor cell morphological maturation; however, these cells survived as round cell bodies without processes for at least 10 days in the presence of RPE-CM and expressed opsin during this period. Conditioned medium from the flat-cell monolayers did not support photoreceptor differentiation or their survival. However, the presence of flat cells was a requisite to achieve any neurite outgrowth even in the presence of RPE-CM. In the absence of RPE-CM, neither photoreceptors nor flat cells survived or proliferated. Heat and trypsin treatment of the RPE-CM abolished all its growth-supporting activities which indicates its proteinaceous nature. This represents the first time in vitro that an RPE-derived factor(s) has been shown to be responsible for *photoreceptor* *cell* *survival* and differentiation.

14/3,AB/12
DIALOG(R)File 155:MEDLINE(R)

07166640 92094687 PMID: 1721739
Transplantation to the diseased and damaged retina.
Sheedlo H J; Gaur V; Li L X; Seaton A D; Turner J E
Dept of Neurobiology and Anatomy, Bowman Gray School of Medicine, Wake Forest University, Winston-Salem, NC 27103.
Trends in neurosciences (ENGLAND) Aug 1991, 14

(8) p347-50, ISSN 0166-2236 Journal Code: 7808616
Document type: Journal Article; Review; Review, Tutorial
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Retinas of Royal College of Surgeons (RCS) dystrophic rats undergo a dramatic loss of photoreceptor cells as a result of defective retinal pigment epithelial (RPE) cells. These retinas are therefore a valuable model in the investigation of the role of the RPE on *photoreceptor*- *cell* *survival* and development. Also, rat retinas damaged by excessive light serve as a suitable environment to study survival of transplanted photoreceptor cells. Even though photoreceptor cells are lost in these retinas, a normal inner retinal structure is retained. Both models have recently been used in successful RPE-cell and/or photoreceptor-cell transplantation studies designed to replace defective or lost cells due to retinal disease or damage. These new approaches in the field of retinal transplantation offer unique and novel opportunities for the development of possible therapeutic strategies in human eye disease, and for improving our understanding of the normal relationships between retinal cells.

14/3,AB/13
DIALOG(R)File 155:MEDLINE(R)

07135494 92070150 PMID: 1959383
Effects of macrophage and retinal pigment epithelial cell transplants on photoreceptor cell rescue in RCS rats.
Li L X; Sheedlo H J; Gaur V; Turner J E
Department of Neurobiology and Anatomy, Bowman Gray School of Medicine, Wake Forest University, Winston-Salem, NC.
Current eye research (ENGLAND) Oct 1991, 10 (10) p947-58, ISSN 0271-3683 Journal Code: 8104312
Contract/Grant No.: EY04337; EY; NEI
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
The effects of macrophage transplants on *photoreceptor* *cell* *survival* in retinas of Royal College of Surgeons (RCS) dystrophic rats were contrasted with RPE-cell transplants, sham-injection and surgical controls. The effects of these different treatments on the thickness and total area of the outer nuclear layer (ONL) were evaluated by light and electron microscopy at 1, 2 and 5 months after transplantation or surgical manipulations. Macrophage transplants into dystrophic retinas, although significantly reducing the debris zone thickness (p less than 0.01), had little effect on *photoreceptor* *cell* *survival* (2-3 cells thick ONL) after two months. In contrast, two months after RPE-cell transplantation, retinas exhibited an 8-10 cell thick ONL. Also, inner and outer segments of rescued photoreceptor cells were present, especially in areas directly beneath RPE-cell transplants. At the same time period, retinas injected with saline had a 2-3 cell thick ONL with no organized inner or outer segments. Furthermore, the affected ONL area in macrophage-transplanted or saline-injected retinas was significantly smaller than that seen in RPE-cell transplanted retinas (p less than 0.0001). Surviving photoreceptor cells were found only in the RPE-cell transplanted retinas five months after treatment. No effect on *photoreceptor* *cell* *survival* was seen in saline-injected, needle-inserted or incision-only retinas. Thus, transplantation of healthy RPE cells is an effective long-term therapeutic approach to correct the genetic defect in retinas of RCS dystrophic rats.

? s cornelia()de()lange
238 CORNELIA
1862402 DE
1147 LANGE
S15 227 CORNELIA()DE()LANGE
? s cornelia()de()lange()syndrome
238 CORNELIA
1862402 DE
1147 LANGE
473381 SYNDROME
S16 184 CORNELIA()DE()LANGE()SYNDROME
? s s15 and expression
227 S15
537170 EXPRESSION
S17 8 S15 AND EXPRESSION
? t s17/3,ab/all

17/3,AB/1
DIALOG(R)File 155:MEDLINE(R)

10397890 99408239 PMID: 10480362
Genomic organisation of the human chordin gene and mutation screening of candidate *Cornelia* *de* *Lange* syndrome genes. Smith M; Herrell S; Lusher M; Lako L; Simpson C; Wiestner A; Skoda R; Ireland M; Strachan T
Human Molecular Genetics Unit, School of Biochemistry and Genetics, University of Newcastle upon Tyne, UK.
Human genetics (GERMANY) Jul-Aug 1999, 105 (1-2) p104-11, ISSN 0340-6717 Journal Code: 7613873
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
We have determined the genomic organisation of the human chordin gene, CHRD, and have shown that it maps within a gene cluster at 3q27 containing THPO

(thrombopoietin), CLCN2 (a voltage-gated chloride-channel gene) and EIF4G1 (a eukaryotic translation-initiation-factor-gamma gene). The CHRD and THPO genes are very close neighbours and are transcribed from opposing DNA strands from promoters that are spaced less than 2 kb apart. We considered that the CHRD gene and the chordin-regulating GSC (goosecoid) gene could be candidate genes for *Cornelia* *de* *Lange* syndrome (CDLS), a developmental malformation syndrome which is primarily characterised by mental handicap, growth retardation, distinctive facial features and limb-reduction defects. CDLS patients typically occur as sporadic cases, but several reports have suggested dominant inheritance. The candidacy of the CHRD and GSC genes was supported by several lines of evidence: prior evidence for a CDLS gene at 3q26.3-q27; a report suggesting a significant association between CDLS and thrombocytopenia; suspected genetic heterogeneity in CDLS; location of the GSC gene in close proximity to a 14q32 breakpoint detected in a CDLS patient with a balanced de novo translocation; known regulation of chordin *expression* by goosecoid; and the pattern of embryonic *expression* of the mouse GSC gene. Another candidate gene at 3q27, SOX2, was also considered because of its suspected role as a transcription factor in early development and because of known examples of SOX genes that are loci for dominantly inherited developmental disorders. However, mutation screening failed to identify CDLS patient-specific mutations in CHRD, GSC or SOX2.

17/3,AB/2
DIALOG(R)File 155:MEDLINE(R)

09830911 98277817 PMID: 9615583
Cecal volvulus as a complication in *Cornelia* *de* *Lange* syndrome. A case report and literature review]
Cokal volvulus som komplikasjon ved Cornelia de Langes syndrom. En kasuistikk med gjennomgang av litteraturen.
Holthusen J; Rottingen J A
Kirurgisk avdeling Stokmarknes sykehus.
Tidsskrift for den Norske laegeforening (NORWAY) Apr 20 1998, 118 (10) p1559-60, ISSN 0029-2001 Journal Code: 0413423
Document type: Journal Article ; English Abstract
Languages: NORWEGIAN
Main Citation Owner: NLM
Record type: Completed
Colonic volvulus in children is a rare, but serious and important differential diagnosis in acute abdominal illness. Our patient with *Cornelia* *de* *Lange*'s syndrome, was admitted with an acute onset of abdominal pain and in a critical condition. Explorative laparotomy revealed a caecal volvulus with necrosis of the distal ileum, caecum and proximal colon. The syndrome is characterized by typical facial *expression* , both growth and mental retardation, and various gastrointestinal and cardiac anomalies. Predisposing factors contributing to volvulus in this syndrome are mental retardation and a higher incidence of malrotation and nonfixation of the caecum and ascending colon. The parents of children with *Cornelia* *de* *Lange*'s syndrome should therefore be counselled so that they are able to provide essential information in the event of their children experiencing acute illness.

17/3,AB/3
DIALOG(R)File 155:MEDLINE(R)

09725393 98133920 PMID: 9466998
A new human homeobox gene OGI2X is a member of the most conserved homeobox gene family and is expressed during heart development in mouse. Semina E V; Reiter R S; Murray J C
Departments of Pediatrics and Biological Sciences, The University of Iowa, Iowa City, IA 52242, USA.
Human molecular genetics (ENGLAND) Mar 1998, 7 (3) p415-22, ISSN 0964-6906 Journal Code: 9208958
Contract/Grant No.: DE-08559; DE; NIDCR; DE-09170; DE; NIDCR; DK-25295; DK; NIDDK
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Homeodomain (HD) proteins are transcription regulators controlling a variety of cell fates. The HD region characterizing this protein family is a domain of 60 amino acid residues that recognizes and binds a site in the regulatory region of the target gene. It has been suggested that regions outside the HD may determine the specific functions of the various HD proteins by forming additional contacts with DNA sequences or by interactions with other proteins. We have identified a 14 amino acid motif within the C-terminal region of the protein encoded by the RIEG1 gene that is conserved among several HD proteins. Overlapping *expression* of the genes encoding these proteins during craniofacial development suggested that they might interact with a common factor. In order to identify additional genes possessing this motif we screened a human craniofacial cDNA library with oligoprobes. A novel gene was identified, exhibiting the most homology to murine Og12x (formerly OG12) and the recently reported human SHOX gene. Human OG12X and murine Og12x are highly homologous and the OG12X and Og12x proteins are 100% identical. In situ hybridization on mouse embryos ranging from 9 to 16 days post-coitum localized murine Og12x mRNA in the heart, otic region, maxillary and mandibular components of the first branchial arch, nasal processes,

eyelid, midbrain, medulla oblongata, limbs, dorsal root ganglia and genital tubercle. OG12X was mapped to human chromosome 3q22-26 and murine Og12x to the syntenic region on mouse chromosome 3. Based upon the *expression* pattern of its mouse cognate, OG12X represents a candidate for the blepharophimosis (BPES) and *Cornelia* *de* *Lange* syndromes previously mapped to this region.

17/3,AB/4
DIALOG(R)File 155:MEDLINE(R)

09724515 98151525 PMID: 9482898
SHOT, a SHOX-related homeobox gene, is implicated in craniofacial, brain, heart, and limb development.
Blaschke R J; Monaghan A P; Schiller S; Schechinger B; Rao E; Padilla-Nash H; Ried T; Rappold G A
Institute of Human Genetics, Heidelberg University, Im Neuenheimer Feld 328, 69120 Heidelberg, Germany.
Proceedings of the National Academy of Sciences of the United States of America (UNITED STATES) Mar 3 1998, 95 (5) p2406-11, ISSN 0027-8424 Journal Code: 7505876
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Deletion of the SHOX region on the human sex chromosomes has been shown to result in idiopathic short stature and proposed to play a role in the short stature associated with Turner syndrome. We have identified a human paired-related homeobox gene, SHOT, by virtue of its homology to the human SHOX and mouse OG-12 genes. Two different isoforms were isolated, SHOTa and SHOTb, which have identical homeodomains and share a C-terminal 14-amino acid residue motif characteristic for craniofacially expressed homeodomain proteins. Differences between SHOTa and b reside within the N termini and an alternatively spliced exon in the C termini. In situ hybridization of the mouse equivalent, OG-12, on sections from staged mouse embryos detected highly restricted transcripts in the developing sinus venosus (aorta), female genitalia, diencephalon, mes- and myelencephalon, nasal capsula, palate, eyelid, and in the limbs. SHOT was mapped to human chromosome 3q25-q26 and OG-12 within a syntenic region on chromosome 3. Based on the localization and *expression* pattern of its mouse homologue during embryonic development, SHOT represents a candidate for the *Cornelia* *de* *Lange* syndrome.

17/3,AB/5
DIALOG(R)File 155:MEDLINE(R)

03137674 79211076 PMID: 455352
Typus degenerativus amstelodamensis. The *Cornelia* *de* *Lange* syndrome in 2 children]
Typus degenerativus amstelodamenis. *Cornelia* *de* *lange* syndrom u dvou deti.
Seemanova E; Losan F; Salichova J
Casopis lekar u c eskych (CZECHOSLOVAKIA) Mar 30 1979, 118 (13) p404-7, ISSN 0008-7335 Journal Code: 0004743
Document type: Journal Article ; English Abstract
Languages: CZECH
Main Citation Owner: NLM
Record type: Completed

17/3,AB/6
DIALOG(R)File 155:MEDLINE(R)

02287294 76110708 PMID: 4469431
Example of the *Cornelia* *de* *Lange* syndrome] Un exemple du syndrome de *Cornelia* *de* *Lange*
Bourlond A; Sacre P; Nicolay M
Archives belges de dermatologie (BELGIUM) Oct-Dec 1974, 30 (4) p293-5, ISSN 0301-8636 Journal Code: 0412667
Document type: Journal Article ; English Abstract
Languages: FRENCH
Main Citation Owner: NLM
Record type: Completed

17/3,AB/7
DIALOG(R)File 155:MEDLINE(R)

01421042 72181062 PMID: 5148360
Self-mutilative behavior in the *Cornelia* *de* *Lange* syndrome.
Bryson Y; Sakati N; Nyhan W L; Fish C H
American journal of mental deficiency (UNITED STATES) Nov 1971, 76 (3) p319-24, ISSN 0002-9351 Journal Code: 0372647
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

17/3,AB/8
DIALOG(R)File 155:MEDLINE(R)

00234177 66153849 PMID: 5939543
Dermatological manifestations of the *Cornelia* *de* *Lange* syndrome.
Salazar F N

Archives of dermatology (UNITED STATES) Jul 1966, 94 (1) p38-43, ISSN 0003-987X Journal Code: 0372433
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
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